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(54) **ATTENUATED SWINE INFLUENZA VACCINES AND METHODS OF MAKING AND USE THEREOF**

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(58) **Field of Classification Search**

None

See application file for complete search history.

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(57) **ABSTRACT**

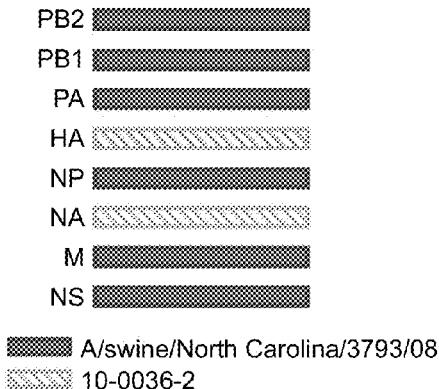
This disclosure provides attenuated swine influenza strains, particularly those produced via a reverse genetics approach, compositions comprising same, and methods of production and use thereof. The attenuated strains are engineered to encode HA proteins having additional glycosylation sites, relative to the HA proteins encoded by the corresponding virulent parental viruses. Advantageously, the attenuated influenza strains may be administered.

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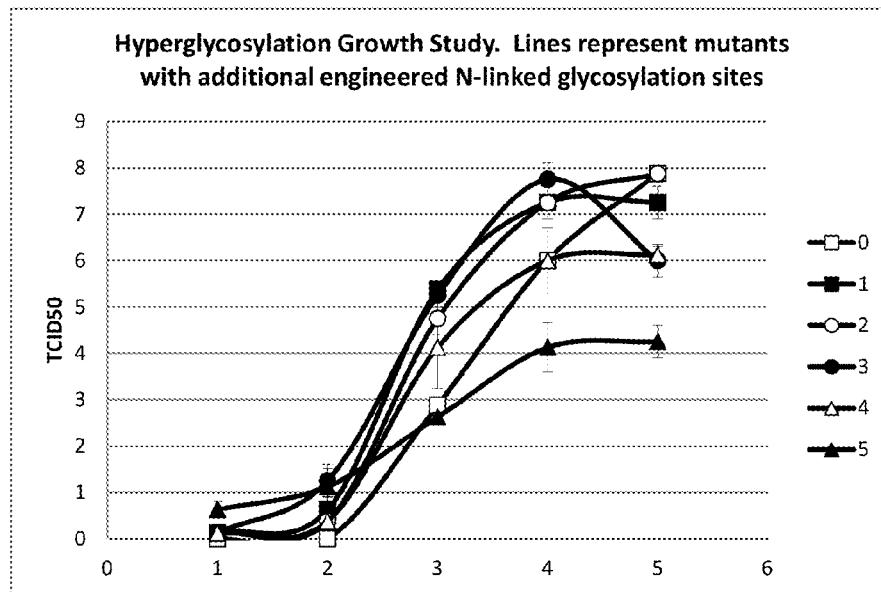
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**FIG. 1**

**Seq 18 = HA (parent); Seq 22 = HA (n+5 mutant)**

<b>SEQ 18</b>	MKVKLMVLLCTFTATYADTICVGYHANNSTDVTVDLKNVTVTHSVNLLEDSHNGKLCL	60
<b>SEQ 22</b>	MKVKLMVLLCTFTATYADTICVGYHANNSTDVTVDLKNVTVTHSVNLLEDSHNGKLCL	60
<b>SEQ 18</b>	LKGIAPLQLGCSVAGWILGNPECELLIS	120
<b>SEQ 22</b>	LKGIAPLQLGCSVAGWILGNPECELLIS	120
<b>SEQ 18</b>	QLSSVSSFKRFEIFPKESSWPNHTVTGVSSSCSHNGKSSFYRNLLWLTVKNGYPNLSKS	180
<b>SEQ 22</b>	QLSSVSSFKRFEIFPKESSWPNHTVTGVSSSCSHNGKSSFYRNLLWLTVKNGYPNLSKS	180
<b>SEQ 18</b>	YTNNKEKEVILVIWGVHHPSNIGDQRALYHTENAYVSVSSHYSRRTPEIAKRPKVRNQE	240
<b>SEQ 22</b>	YTNNKEKEVILVIWGVHHPSNIGDQRALYHTENAYVSVSSHYSRRTPEIAKRPKVRNQE	240
<b>SEQ 18</b>	GRINYYWTLEPGDTIIFEANGNLIAPRYAFELSKGFEGSGIITSNA	300
<b>SEQ 22</b>	GRINYYWTLEPGDTIIFEANGNLIAPRYAFELSKGFQSCIITSNA	300
<b>SEQ 18</b>	AINSSLPPQNVPVTIGECPKYVKSAKLRMVTGLRNTPSIQSRGLFGAIAGFIEGGWTGM	360
<b>SEQ 22</b>	AINSSLPPQNVPVTIGECPKYVKSAKLRMVTGLRNTPSIQSRGLFGAIAGFIEGGWTGM	360
<b>SEQ 18</b>	VDGWYGYHHQNEQGSGYAADQQSTQNAINGITNKVNNSIEKMNTQFTAVGKEFNKLERRM	420
<b>SEQ 22</b>	VDGWYGYHHQNEQGSGYAADQQSTQNAINGITNKVNNSIEKMNTQFTAVGKEFNKLERRM	420
<b>SEQ 18</b>	ENLNKKVDDGFLDIWTYNAELLVLLENERTLDFHDSNVKNLYEKVSQLKNNAKEIGNGC	480
<b>SEQ 22</b>	ENLNKKVDDGFLDIWTYNAELLVLLENERTLDFHDSNVKNLYEKVSQLKNNAKEIGNGC	480
<b>SEQ 18</b>	FEFYHKCNDECMEVKNGTYDYPKYSEESKLNREKIDGVKLESMSGVYNILAIYSTVASSL	540
<b>SEQ 22</b>	FEFYHKCNDECMEVKNGTYDYPKYSEESKLNREKIDGVKLESMSGVYNILAIYSTVASSL	540
<b>SEQ 18</b>	VLLVSLGAISFWMCNSGSLQCRICI	565
<b>SEQ 22</b>	VLLVSLGAISFWMCNSGSLQCRICI	565

**FIG. 2**



	Rescued virus	additional glycosylation sites (n)
0	RG parent (10-0036-2)	0
1	S71N	1
2	S71N, K90N	2
3	S71N, K90N, L173T	3
4	S71N, K90N, L173T, P287T	4
5	S71N, K90N, L173T, P287T, K294T	5

**FIG. 3**

SEQ ID NO	TYPE	DESCRIPTION
1	DNA	PB2 gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
2	PRT	PB2 predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
3	DNA	PB1 gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
4	PRT	PB1 predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
5	DNA	PA gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
6	PRT	PA predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
7	DNA	HA gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
8	PRT	HA predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
9	DNA	NP gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
10	PRT	NP predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
11	DNA	NA gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
12	PRT	NA predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
13	DNA	M gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
14	PRT	M predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
15	DNA	NS gene of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
16	PRT	NS predicted AA of Influenza A/swine/NorthCarolina/3793/08(H1N1) - 10-0036-2
17	DNA	HA gene of Influenza 10-0036-2 (n+0, parent) - RG 10-0036-2
18	PRT	HA predicted AA of Influenza 10-0036-2 (n+0, = parent) - RG 10-0036-2
19	DNA	NA gene of Influenza 10-0036-2 (n+0, parent) - RG 10-0036-2
20	PRT	NA predicted AA of Influenza 10-0036-2 (n+0, = parent) - RG 10-0036-2
21	DNA	HA gene of Influenza 10-0036-2 (n+5, relative to RG 10-0036-2)
22	PRT	HA predicted AA of Influenza 10-0036-2 (n+5, relative to RG 10-0036-2)
23	DNA	HA gene of Influenza 10-0036-2 (n+4, relative to RG 10-0036-2)
24	PRT	HA predicted AA of Influenza 10-0036-2 (n+4, relative to RG 10-0036-2)
25	DNA	HA gene of Influenza 10-0036-2 (n+3, relative to RG 10-0036-2)
26	PRT	HA predicted AA of Influenza 10-0036-2 (n+3, relative to RG 10-0036-2)
27	DNA	HA gene of Influenza 10-0036-2 (n+2, relative to RG 10-0036-2)
28	PRT	HA predicted AA of Influenza 10-0036-2 (n+2, relative to RG 10-0036-2)
29	DNA	HA gene of Influenza 10-0036-2 (n+1, relative to RG 10-0036-2)
30	PRT	HA predicted AA of Influenza 10-0036-2 (n+1, relative to RG 10-0036-2)

**FIG. 4**

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**ATTENUATED SWINE INFLUENZA  
VACCINES AND METHODS OF MAKING  
AND USE THEREOF**

**INCORPORATION BY REFERENCE**

This application claims priority to provisional application U.S. Ser. No. 61/672,398, filed on Jul. 17, 2012, and incorporated by reference herein in its entirety.

**FIELD OF THE INVENTION**

The present invention relates generally to attenuated viral vaccines, particularly those providing broad, safe, and effective protection to porcines against infections/disease caused by swine influenza. The invention further relates to methods of producing the attenuated viruses, and to the identification of nucleic acid variations that are associated with decreased virulence of the attenuated virus.

The invention accordingly relates to immunogenic or vaccine compositions comprising the viruses of the invention; e.g., live attenuated virus. The viruses also could be inactivated in the compositions; but it may be advantageous that the viruses are live attenuated swine influenza. The invention therefore further relates to methods for preparing and/or formulating such compositions; e.g., culturing or growing or propagating the viruses on or in suitable medium, harvesting the viruses, optionally inactivating the viruses, and optionally admixing with a suitable veterinarianally or pharmaceutically acceptable carrier, excipient, diluent or vehicle and/or an adjuvant and/or stabilizer. Thus, the invention also relates to the use of the viruses in formulating such compositions.

**BACKGROUND OF THE INVENTION**

Influenza viruses possess a number of mechanisms to elude host humoral immunity against hemagglutinin (HA) that mediates viral entry. Reassortment of genome segments can lead to antigenic shift while gradual accumulation of mutations leads to genetic drift (Desselberger U, et al. 1978, Schild G, et al. 1974). In addition to mutations in the antigenic epitopes of the HA that can impact the ability of pre-existing antibodies to recognize mutant HA, mutations in N-linked glycosylation of HA can promote viral evasion of antibody recognition by altering the oligosaccharide layer surrounding the HA (Schulze I T, 1997). Glycan residues can restrict the binding of some antibodies to their epitopes, leading to loss of antibody recognition and immunogenicity in a phenomenon known as glycan shielding (Wanzeck K, et al. 2011, Wei C-J, et al. 2010). Additionally, mutation and genetic drift preferentially occur at positions in the HA globular head that are not protected by glycans (Das S R, et al. 2010). Glycan residues also influence receptor binding and consequently can affect viral replication kinetics.

The evolution of influenza viruses frequently includes modification of the number and position of glycosylation sites (Long J, et al. 2011). For H3N2 viruses, the number of glycosylation sites has increased from two to ten over the last 40 years (4). However, increasingly glycosylated HA has been correlated with decreased virulence in mice and reduced viral fitness (Das S R, et al. 2011, Vigerust D J, et al. 2007). While increasing glycosylation can shield HA from neutralizing antibodies, the viral affinity for cellular receptors is obligatorily decreased (Abe Y, et al. 2004, Das S R, et al. 2011). Also, compensatory mutations in either the HA or neuraminidase (NA) are required to balance receptor binding and release activities (Wagner R, et al. 2000). These compen-

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satory mutations in NA have been associated with the acquisition of natural resistance to NA inhibitors (Hensley S E, et al. 2011).

Numerous genetic and antigenically distinct lineages of swine influenza virus circulate concurrently in pigs, including  $\beta$ ,  $\gamma$  and  $\delta$ -cluster H1N1 and H1N2 viruses, as well as the H3N2 subtype (Lorusso A, et al. 2012). The  $\delta$ -cluster was originally subdivided into two subclusters ( $\delta$ -I and  $\delta$ -II), however, more recent work demonstrated five distinct genetic subclusters ( $\delta$ -A,  $\delta$ -B,  $\delta$ -C,  $\delta$ -D,  $\delta$ -E) representing at least three antigenically distinct groups (Hause B M, et al. 2011, Vincent A L, et al. 2009).

While killed viral vaccines are efficacious when the vaccine strains match the field challenge virus, the rapid mutation rate of influenza requires frequent strain changes to ensure genetic and antigenic match between the vaccine strains and circulating viruses. Additionally, the multiple subtypes and lineages co-circulating in swine frequently require five or more vaccine strains in order to include representatives of each type. A single live vaccine that elicits broad spectrum immunity offer a significant advance in swine health programs.

**SUMMARY OF THE INVENTION**

An object of this invention is to provide attenuated vaccines as well as methods for treatment and prophylaxis of infection by swine influenza.

In an embodiment, the vaccines comprise attenuated influenza viruses, which have been modified to express HA genes having additional glycosylation sites relative to their parental strain.

In a particular embodiment, the HA genes and/or gene products have the same modifications as are present between SEQ ID NOs:17 and 21; SEQ ID NOs:17 and 23; SEQ ID NOs:18 and 22; or SEQ ID NOs:18 and 24. In yet another embodiment, the HA genes and/or gene products have at least 1 out of the 5 modifications. In a more particular embodiment, the HA genes and/or gene products have 4 or 5 of the modifications, results in +4 and +5 glycosylation sites, relative to the parent HA gene products.

Another object of this invention is to provide cDNA and/or plasmids for use in a reverse genetics system for producing attenuated influenza according to the instant disclosure. In an embodiment, at least one of the cDNA and/or plasmids comprises a HA sequence having increased number of glycosylation sites relative to the sequence as set forth in SEQ ID NO:18.

In a particular embodiment, the HA sequence is as set forth in SEQ ID NOs:21 or 23.

The present invention further relates to new attenuated strains of Influenza, which provide safe, effective, and broad protective immunity. Relative to a parent Influenza strain, the attenuated strains may have additional glycosylation sites and/or putative glycosylation sites encoded by their HA genes, whose presence is associated with reduced virulence.

Thus, the invention provides a mutant virus comprising a mutation(s) in one or more nucleic acids sequences, relative to the wild type/parental virus, which renders the mutant virus attenuated, relative to the parent virus, which parent virus comprises nucleic acids encoding a wild type HA protein. As defined herein, the "wild type" HA protein is one having the same number of glycosylation sites and/or putative glycosylation sites relative to the sequences as set forth in SEQ ID NO:18.

In a particular embodiment, the mutant virus comprises vRNA nucleic acid sequences which correspond to (i.e. are

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reverse complementary and have uracils in place of thymines) the DNA sequences set forth in SEQ ID NOs:21 or 23, which encode for peptides as set forth in SEQ ID NOs:22 or 24, respectively, and which cause the mutant virus to be attenuated/non-virulent, relative to the virulent wild type/parental virus.

As defined herein, the term "gene" will be used in a broad sense, and shall encompass both coding and non-coding sequences (i.e. upstream and downstream regulatory sequences, promoters, 5'/3' UTR, introns, and exons). Where reference to only a gene's coding sequence is intended, the term "gene's coding sequence" or "CDS" will be used interchangeably throughout this disclosure. When a specific nucleic acid is discussed, for example, the sequence as set forth in SEQ ID NO:17 (the DNA sequence equivalent of parental virus cRNA "sense" strand), the skilled person will instantly be in possession of all derivable forms of that sequence (mRNA, vRNA, cRNA, DNA, protein, etc.). For example, the influenza virus is a negative single strand RNA virus (ssRNA). To replicate, its negative ssRNA (defined herein as "vRNA") must be transcribed to positive or sense RNA (defined herein as "cRNA"). Host cell machinery is co-opted to use the cRNA to produce the viral proteins and vRNA. A skilled person using the well-known genetic code can routinely derive from a DNA sequence the vRNA, cRNA, and peptide sequences.

In a particular embodiment, the attenuated vaccines comprise an adjuvant. The adjuvant may be any substance which increases and/or augments the elicited immune response, as compared to attenuated vaccine alone. Mucosal adjuvants, including chitosans and derivatives thereof, are particularly useful for the disclosed oral attenuated vaccines.

The invention further provides methods for inducing an immunological (or immunogenic) or protective response against Influenza, as well as methods for preventing or treating Influenza, or disease state(s) caused by Influenza, comprising administering the attenuated virus, or a composition comprising the attenuated virus to animals in need thereof.

Kits comprising at least the attenuated Influenza strain and instructions for use are also provided.

These and other embodiments are disclosed or are obvious from and encompassed by, the following Detailed Description.

## BRIEF DESCRIPTION OF DRAWINGS

A full and enabling disclosure of the present invention, including the best mode thereof, to one of ordinary skill in the art, is set forth more particularly in the remainder of the specification, including reference to the accompanying figures, wherein:

FIG. 1 presents the genotype of viruses created using reverse genetics;

FIG. 2 is an amino acid alignment between SEQ ID NO:18 (HA protein of parent influenza virus) and SEQ ID NO:22 (HA protein of n+5 glycosylation mutant);

FIG. 3 is a graph of the growth in cultured cells of parent and glycosylation mutant influenza viruses harboring between 1 and 5 additional glycosylation sites in their HA gene, relative to parent;

FIG. 4 is a table listing the SEQ ID of this disclosure.

## DETAILED DESCRIPTION OF THE INVENTION

The present invention provides nucleotide sequences and genes involved in the attenuation of a microorganism, such as virus, for instance, Influenza, products (e.g., proteins, anti-

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gens, immunogens, epitopes) encoded by the nucleotide sequences, methods for producing such nucleotide sequences, products, micro-organisms, and uses therefore, such as for preparing vaccine or immunogenic compositions or for eliciting an immunological or immune response or as a vector, e.g., as an expression vector (for instance, an in vitro or in vivo expression vector).

Mutations introduced into nucleotide sequences and genes of micro-organisms produce novel and nonobvious attenuated mutants. These mutants are useful for the production of live attenuated immunogenic compositions or live attenuated vaccines having a high degree of immunogenicity.

Identification of the mutations provides novel and nonobvious nucleotide sequences and genes, as well as novel and nonobvious gene products encoded by the nucleotide sequences and genes.

In an embodiment, the invention provides an attenuated hyperglycosylated swine influenza strain capable of providing a safe and effective immune response in porcine against influenza or diseases caused by influenza. In one embodiment, the strain may encode an HA gene having at least 1 additional glycosylation site relative to virulent parental strain.

In another embodiment, the attenuated strain may encode an HA gene having 4 or 5 additional glycosylation sites relative to virulent parental strain. In a particular embodiment, the strain glycosylation sites are selected from S71N, K90N, L173T, P287T, and K294T, with the locations of the Amino Acid changes being based upon an HA gene having the sequence as set forth in SEQ ID NO:18.

In one embodiment, the HA protein produced by the attenuated influenza strain has the sequence as set forth in SEQ ID NO:22 or a sequence with at least 90% homology to SEQ ID NO:22 provided the following locations have the following amino acids: 71N, 90N, 173T, 287T, and 294T.

In another embodiment, the attenuated influenza produces an HA protein having the sequence as set forth in SEQ ID NO:24 or a sequence with at least 90% homology to SEQ ID NO:24 provided the following locations have the following amino acids: 71N, 90N, 173T, 287T.

In another aspect, the invention provides immunological composition comprising attenuated influenza strains, which encode hyperglycosylated HA proteins. In one embodiment, the compositions may further comprise a pharmaceutically or veterinary acceptable vehicle, diluent or excipient.

In an embodiment, the composition provides a protective immune response in porcine against virulent swine influenza challenge. In some embodiments, the composition further comprises at least one additional antigen associated with a pathogen other than swine influenza.

In another embodiment, the at least one additional antigen is selected from *M. hyo*, PCV2, PRRSV, SIV or other pathogen capable of infecting and causing illness or susceptibility to illness in a porcine, or combinations thereof.

In an embodiment, the invention provides methods of vaccinating an animal comprising at least one administration of the compositions comprising sequences encoding hyperglycosylated influenza HA proteins. In another embodiment, the porcine is a sow from about 3 weeks to about 6 weeks pre-farrowing. In yet another embodiment, the resulting piglets may have a reduced morbidity and/or mortality as compared to piglets coming from unvaccinated sows.

In another embodiment, the invention provides a composition comprising a plurality of vectors for production of attenuated swine influenza including a vector comprising a promoter operably linked to an influenza virus HA cDNA, wherein the HA cDNA encodes additional glycosylation sites

relative to an HA encoded by a virulent parent swine influenza strain. In one particular embodiment, the additional glycosylation sites are selected from S71N, K90N, L173T, P287T, and K294T, and the location of the Amino Acid changes is based upon an HA gene having the sequence as set forth in SEQ ID NO:18.

In an embodiment, the HA cDNA for producing attenuated influenza encodes the protein as set forth in SEQ ID NO:22. In another embodiment, HA cDNA encodes the protein as set forth in SEQ ID NO:24.

In an embodiment, the invention provides a method to prepare influenza virus, comprising: contacting a cell with one of the inventive compositions in an amount effective to yield infectious influenza virus. In one embodiment, the method further comprises isolating the virus.

In another embodiment, the invention provides a method to prepare a gene delivery vehicle, comprising: contacting cells with the inventive composition in an amount effective to yield influenza virus, and isolating the virus. The invention further provides a cell contacted with the inventive composition.

In an embodiment, the invention provides a vertebrate cell comprising a plurality of vectors for production of attenuated swine influenza including a vector comprising a promoter operably linked to an influenza virus HA cDNA, wherein the HA cDNA encodes additional glycosylation sites relative to an HA encoded by a virulent parent swine influenza strain.

The invention further encompasses gene products, which provide antigens, immunogens and epitopes, and are useful as isolated gene products.

Such isolated gene products, as well as epitopes thereof, are also useful for generating antibodies, which are useful in diagnostic applications.

Such gene products, which can provide or generate epitopes, antigens or immunogens, are also useful for immunogenic or immunological compositions, as well as vaccines.

In an aspect, the invention provides viruses containing an attenuating mutation in a nucleotide sequence or a gene wherein the mutation modifies the biological activity of a polypeptide or protein encoded by a gene, resulting in attenuated virulence of the virus.

In particular, the present invention encompasses attenuated swine influenza strains and vaccines comprising the same, which elicit an immunogenic response in an animal, particularly the attenuated swine influenza strains that elicit, induce or stimulate a response in a porcine.

Particular swine influenza attenuated strains of interest have mutations in genes, relative to wild type virulent parent strain, which are associated with virulence. It is recognized that, in addition to strains having the disclosed mutations, attenuated strains having any number of mutations in the disclosed virulence genes can be used in the practice of this invention.

In another aspect, the novel attenuated swine influenza strains are formulated into safe, effective vaccine against swine influenza and infections/diseases cause by swine influenza.

In an embodiment, the swine influenza vaccines further comprise an adjuvant. In a particular embodiment, the adjuvant is a mucosal adjuvant, such as chitosan, methylated chitosan, trimethylated chitosan, or derivatives or combinations thereof.

In an embodiment, the adjuvant comprises whole bacteria and/or viruses, including *H. parasuis*, clostridium, swine influenza virus (SIV), porcine circovirus (PCV), porcine reproductive and respiratory syndrome virus (PRRSV), *Mannheimia*, *Pasteurella*, *Histophilus*, *Salmonella*, *Escherichia coli*, or combinations and/or variations thereof. In several

embodiments, the adjuvant increases the animal's production of IgM, IgG, IgA, and/or combinations thereof.

By "antigen" or "immunogen" means a substance that induces a specific immune response in a host animal. The antigen may comprise a whole organism, killed, attenuated or live; a subunit or portion of an organism; a recombinant vector containing an insert with immunogenic properties; a piece or fragment of DNA capable of inducing an immune response upon presentation to a host animal; a polypeptide, an epitope, a haptan, or any combination thereof. Alternately, the immunogen or antigen may comprise a toxin or antitoxin.

The terms "protein", "peptide", "polypeptide" and "polypeptide fragment" are used interchangeably herein to refer to polymers of amino acid residues of any length. The polymer can be linear or branched, it may comprise modified amino acids or amino acid analogs, and it may be interrupted by chemical moieties other than amino acids. The terms also encompass an amino acid polymer that has been modified naturally or by intervention; for example disulfide bond formation, glycosylation, lipidation, acetylation, phosphorylation, or any other manipulation or modification, such as conjugation with a labeling or bioactive component.

The term "immunogenic or antigenic polypeptide" as used herein includes polypeptides that are immunologically active in the sense that once administered to the host, it is able to evoke an immune response of the humoral and/or cellular type directed against the protein. Preferably the protein fragment is such that it has substantially the same immunological activity as the total protein. Thus, a protein fragment according to the invention comprises or consists essentially of or consists of at least one epitope or antigenic determinant. An "immunogenic" protein or polypeptide, as used herein, includes the full-length sequence of the protein, analogs thereof, or immunogenic fragments thereof. By "immunogenic fragment" is meant a fragment of a protein which includes one or more epitopes and thus elicits the immunological response described above. Such fragments can be identified using any number of epitope mapping techniques, well known in the art. See, e.g., Epitope Mapping Protocols in Methods in Molecular Biology, Vol. 66 (Glenn E. Morris, Ed., 1996). For example, linear epitopes may be determined by e.g., concurrently synthesizing large numbers of peptides on solid supports, the peptides corresponding to portions of the protein molecule, and reacting the peptides with antibodies while the peptides are still attached to the supports. Such techniques are known in the art and described in, e.g., U.S. Pat. No. 4,708,871; Geysen et al., 1984; Geysen et al., 1986. Similarly, conformational epitopes are readily identified by determining spatial conformation of amino acids such as by, e.g., x-ray crystallography and 2-dimensional nuclear magnetic resonance. See, e.g., Epitope Mapping Protocols, supra. Methods especially applicable to the proteins of *T. parva* are fully described in PCT/US2004/022605 incorporated herein by reference in its entirety.

As discussed herein, the invention encompasses active fragments and variants of the antigenic polypeptide. Thus, the term "immunogenic or antigenic polypeptide" further contemplates deletions, additions and substitutions to the sequence, so long as the polypeptide functions to produce an immunological response as defined herein. The term "conservative variation" denotes the replacement of an amino acid residue by another biologically similar residue, or the replacement of a nucleotide in a nucleic acid sequence such that the encoded amino acid residue does not change or is another biologically similar residue. In this regard, particularly preferred substitutions will generally be conservative in nature, i.e., those substitutions that take place within a family

of amino acids. For example, amino acids are generally divided into four families: (1) acidic—aspartate and glutamate; (2) basic—lysine, arginine, histidine; (3) non-polar—alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tryptophan; and (4) uncharged polar—glycine, asparagine, glutamine, cystine, serine, threonine, tyrosine. Phenylalanine, tryptophan, and tyrosine are sometimes classified as aromatic amino acids. Examples of conservative variations include the substitution of one hydrophobic residue such as isoleucine, valine, leucine or methionine for another hydrophobic residue, or the substitution of one polar residue for another polar residue, such as the substitution of arginine for lysine, glutamic acid for aspartic acid, or glutamine for asparagine, and the like; or a similar conservative replacement of an amino acid with a structurally related amino acid that will not have a major effect on the biological activity. Proteins having substantially the same amino acid sequence as the reference molecule but possessing minor amino acid substitutions that do not substantially affect the immunogenicity of the protein are, therefore, within the definition of the reference polypeptide. All of the polypeptides produced by these modifications are included herein. The term “conservative variation” also includes the use of a substituted amino acid in place of an unsubstituted parent amino acid provided that antibodies raised to the substituted polypeptide also immunoreact with the unsubstituted polypeptide.

The term “epitope” refers to the site on an antigen or hapten to which specific B cells and/or T cells respond. The term is also used interchangeably with “antigenic determinant” or “antigenic determinant site”. Antibodies that recognize the same epitope can be identified in a simple immunoassay showing the ability of one antibody to block the binding of another antibody to a target antigen.

An “immunological response” to a composition or vaccine is the development in the host of a cellular and/or antibody-mediated immune response to a composition or vaccine of interest. Usually, an “immunological response” includes but is not limited to one or more of the following effects: the production of antibodies, B cells, helper T cells, and/or cytotoxic T cells, directed specifically to an antigen or antigens included in the composition or vaccine of interest. Preferably, the host will display either a therapeutic or protective immunological response such that resistance to new infection will be enhanced and/or the clinical severity of the disease reduced. Such protection will be demonstrated by either a reduction or lack of symptoms and/or clinical disease signs normally displayed by an infected host, a quicker recovery time and/or a lowered viral titer in the infected host.

By “animal” is intended mammals, birds, and the like. Animal or host as used herein includes mammals and human. The animal may be selected from the group consisting of equine (e.g., horse), canine (e.g., dogs, wolves, foxes, coyotes, jackals), feline (e.g., lions, tigers, domestic cats, wild cats, other big cats, and other felines including cheetahs and lynx), ovine (e.g., sheep), bovine (e.g., cattle), porcine (e.g., pig), avian (e.g., chicken, duck, goose, turkey, quail, pheasant, parrot, finches, hawk, crow, ostrich, emu and cassowary), primate (e.g., prosimian, tarsier, monkey, gibbon, ape), ferrets, seals, and fish. The term “animal” also includes an individual animal in all stages of development, including newborn, embryonic and fetal stages.

Unless otherwise explained, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. The singular terms “a”, “an”, and “the” include plural referents unless context clearly indicates oth-

erwise. Similarly, the word “or” is intended to include “and” unless the context clearly indicate otherwise.

It is noted that in this disclosure and particularly in the claims and/or paragraphs, terms such as “comprises”, “comprised”, “comprising” and the like can have the meaning attributed to it in U.S. Patent law; e.g., they can mean “includes”, “included”, “including”, and the like; and that terms such as “consisting essentially of” and “consists essentially of” have the meaning ascribed to them in U.S. Patent law, e.g., they allow for elements not explicitly recited, but exclude elements that are found in the prior art or that affect a basic or novel characteristic of the invention.

#### Compositions

The present invention relates to a swine influenza vaccine or composition which may comprise an attenuated swine influenza strain and a pharmaceutically or veterinarily acceptable carrier, excipient, or vehicle, which elicits, induces or stimulates a response in an animal.

The term “nucleic acid” and “polynucleotide” refers to RNA or DNA that is linear or branched, single or double stranded, or a hybrid thereof. The term also encompasses RNA/DNA hybrids. The following are non-limiting examples of polynucleotides: a gene or gene fragment, exons, introns, mRNA, tRNA, rRNA, ribozymes, cDNA, recombinant polynucleotides, branched polynucleotides, plasmids, vectors, isolated DNA of any sequence, isolated RNA of any sequence, nucleic acid probes and primers. A polynucleotide may comprise modified nucleotides, such as methylated nucleotides and nucleotide analogs, uracyl, other sugars and linking groups such as fluororibose and thiolate, and nucleotide branches. The sequence of nucleotides may be further modified after polymerization, such as by conjugation, with a labeling component. Other types of modifications included in this definition are caps, substitution of one or more of the naturally occurring nucleotides with an analog, and introduction of means for attaching the polynucleotide to proteins, metal ions, labeling components, other polynucleotides or solid support. The polynucleotides can be obtained by chemical synthesis or derived from a microorganism.

The term “gene” is used broadly to refer to any segment of polynucleotide associated with a biological function. Thus, genes include introns and exons as in genomic sequence, or just the coding sequences as in cDNAs and/or the regulatory sequences required for their expression. For example, gene also refers to a nucleic acid fragment that expresses mRNA or functional RNA, or encodes a specific protein, and which includes regulatory sequences.

An “isolated” biological component (such as a nucleic acid or protein or organelle) refers to a component that has been substantially separated or purified away from other biological components in the cell of the organism in which the component naturally occurs, for instance, other chromosomal and extra-chromosomal DNA and RNA, proteins, and organelles. Nucleic acids and proteins that have been “isolated” include nucleic acids and proteins purified by standard purification methods. The term also embraces nucleic acids and proteins prepared by recombinant technology as well as chemical synthesis.

The term “conservative variation” denotes the replacement of an amino acid residue by another biologically similar residue, or the replacement of a nucleotide in a nucleic acid sequence such that the encoded amino acid residue does not change or is another biologically similar residue. In this regard, particularly preferred substitutions will generally be conservative in nature, as described above.

The term "recombinant" means a polynucleotide with semisynthetic, or synthetic origin which either does not occur in nature or is linked to another polynucleotide in an arrangement not found in nature.

"Heterologous" means derived from a genetically distinct entity from the rest of the entity to which it is being compared. For example, a polynucleotide may be placed by genetic engineering techniques into a plasmid or vector derived from a different source, and is a heterologous polynucleotide. A promoter removed from its native coding sequence and operatively linked to a coding sequence other than the native sequence is a heterologous promoter.

The polynucleotides of the invention may comprise additional sequences, such as additional encoding sequences within the same transcription unit, controlling elements such as promoters, ribosome binding sites, 5'UTR, 3'UTR, transcription terminators, polyadenylation sites, additional transcription units under control of the same or a different promoter, sequences that permit cloning, expression, homologous recombination, and transformation of a host cell, and any such construct as may be desirable to provide embodiments of this invention.

#### Methods of use and Article of Manufacture

The present invention includes the following method embodiments. In an embodiment, a method of vaccinating an animal comprising administering a composition comprising an attenuated swine influenza strain and a pharmaceutical or veterinarianily acceptable carrier, excipient, or vehicle to an animal is disclosed. In one aspect of this embodiment, the animal is a porcine.

In one embodiment of the invention, a prime-boost regimen can be employed, which is comprised of at least one primary administration and at least one booster administration using at least one common polypeptide, antigen, epitope or immunogen. Typically the immunological composition or vaccine used in primary administration is different in nature from those used as a booster. However, it is noted that the same composition can be used as the primary administration and the booster administration. This administration protocol is called "prime-boost".

A prime-boost regimen comprises at least one prime-administration and at least one boost administration using at least one common polypeptide and/or variants or fragments thereof. The vaccine used in prime-administration may be different in nature from those used as a later booster vaccine. The prime-administration may comprise one or more administrations. Similarly, the boost administration may comprise one or more administrations.

The dose volume of compositions for target species that are mammals, e.g., the dose volume of pig or swine compositions, based on viral antigens, is generally between about 0.1 to about 2.0 ml, between about 0.1 to about 1.0 ml, and between about 0.5 ml to about 1.0 ml.

The efficacy of the vaccines may be tested about 2 to 4 weeks after the last immunization by challenging animals, such as porcine, with a virulent strain of swine influenza. Both homologous and heterologous strains are used for challenge to test the efficacy of the vaccine. The animal may be challenged by IM or SC injection, spray, intra-nasally, intra-ocularly, intra-tracheally, and/or orally. Samples from joints, lungs, brain, and/or mouth may be collected before and post-challenge and may be analyzed for the presence of swine influenza-specific antibody.

The compositions comprising the attenuated viral strains of the invention used in the prime-boost protocols are contained in a pharmaceutically or veterinary acceptable vehicle,

diluent or excipient. The protocols of the invention protect the animal from swine influenza and/or prevent disease progression in an infected animal.

The various administrations are preferably carried out 1 to 5 weeks apart. Preferred time interval is 3 to 5 weeks, and optimally 4 weeks according to one embodiment, an annual booster is also envisioned. The animals, for example pigs, may be at least 3-4 weeks of age at the time of the first administration.

10 It should be understood by one of skill in the art that the disclosure herein is provided by way of example and the present invention is not limited thereto. From the disclosure herein and the knowledge in the art, the skilled artisan can determine the number of administrations, the administration route, and the doses to be used for each injection protocol, without any undue experimentation.

Another embodiment of the invention is a kit for performing a method of eliciting or inducing an immunological or protective response against swine influenza in an animal comprising an attenuated swine influenza immunological composition or vaccine and instructions for performing the method of delivery in an effective amount for eliciting an immune response in the animal.

Another embodiment of the invention is a kit for performing a method of inducing an immunological or protective response against swine influenza in an animal comprising a composition or vaccine comprising an attenuated swine influenza strain of the invention, and instructions for performing the method of delivery in an effective amount for eliciting an immune response in the animal.

Yet another aspect of the present invention relates to a kit for prime-boost vaccination according to the present invention as described above. The kit may comprise at least two vials: a first vial containing a vaccine or composition for the prime-vaccination according to the present invention, and a second vial containing a vaccine or composition for the boost-vaccination according to the present invention. The kit may advantageously contain additional first or second vials for additional prime-vaccinations or additional boost-vaccinations.

40 The pharmaceutically or veterinarianily acceptable carriers or vehicles or excipients are well known to the one skilled in the art. For example, a pharmaceutically or veterinarianily acceptable carrier or vehicle or excipient can be a 0.9% NaCl (e.g., saline) solution or a phosphate buffer. Other pharmaceutically or veterinarianily acceptable carrier or vehicle or excipients that can be used for methods of this invention include, but are not limited to, poly-(L-glutamate) or polyvinylpyrrolidone. The pharmaceutically or veterinarianily acceptable carrier or vehicle or excipients may be any compound or combination of compounds facilitating the administration of the vector (or protein expressed from an inventive vector in vitro); advantageously, the carrier, vehicle or excipient may facilitate transfection and/or improve preservation of the vector (or protein). Doses and dose volumes are herein discussed in the general description and can also be determined by the skilled artisan from this disclosure read in conjunction with the knowledge in the art, without any undue experimentation.

45 The immunological compositions and vaccines according to the invention may comprise or consist essentially of one or more adjuvants. Suitable adjuvants for use in the practice of the present invention are (1) polymers of acrylic or methacrylic acid, maleic anhydride and alkenyl derivative polymers, (2) immunostimulating sequences (ISS), such as oligodeoxyribonucleotide sequences having one or more non-methylated CpG units (Klinman et al., 1996; WO98/16247), (3) an oil in water emulsion, such as the SPT emulsion

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described on page 147 of "Vaccine Design, The Subunit and Adjuvant Approach" published by M. Powell, M. Newman, Plenum Press 1995, and the emulsion MF59 described on page 183 of the same work, (4) cationic lipids containing a quaternary ammonium salt, e.g., DDA (5) cytokines, (6) aluminum hydroxide or aluminum phosphate, (7) saponin or (8) other adjuvants discussed in any document cited and incorporated by reference into the instant application, or (9) any combinations or mixtures thereof.

In an embodiment, adjuvants include those which promote improved absorption through mucosal linings. Some examples include MPL, LTK63, toxins, PLG microparticles and several others (Vajdy, M. Immunology and Cell Biology (2004) 82, 617-627). In an embodiment, the adjuvant may be a chitosan (Van der Lubben et al. 2001; Patel et al. 2005; Majithiya et al. 2008; U.S. Pat. No. 5,980,912).

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The invention will now be further described by way of the following non-limiting examples.

## EXAMPLES

### Example 1

#### Construction of Hyperglycosylated Swine Influenza Viruses

##### Materials/Methods.

Clinical samples (nasal swabs or lung tissue) were collected from pigs exhibiting influenza-like illness and were submitted for viral isolation and characterization (routine diagnostic testing). Virus isolation was performed on swine testicle (ST) cells grown in DMEM containing 5% fetal bovine serum at 37° C. with 5% CO<sub>2</sub>. For viral propagation, fetal bovine serum was omitted from the DMEM. 293T and MDCK cells were propagated in DMEM containing 10% fetal bovine serum. RNA was harvested from infected cell culture harvest fluids using the 5× MagMax-96 Viral Isolation Kit (Life Technologies).

Complete viral genomes were amplified using a previously described multisegment reverse transcription PCR method (Zhou B, et al. 2009). Viral cDNA libraries were prepared using the NEBNext Fast DNA Fragmentation and Library Prep Set 4 kit according to the manufacturer's instructions (New England Biolabs) with the exception that the kit adaptors were replaced with barcoded adaptors (Ion Xpress Barcode Adapter 1-16 Kit, Life Technologies). DNA sequencing templates were prepared using the Ion Xpress Template Kit version 2.0 (Life Technologies) and sequenced using an Ion Torrent Personal Genome Machine (Life Technologies). Contigs were assembled using SeqMan NGen software (DNAStar). Contigs encoding full length HA were identified by BLAST analysis. Full length HA DNA sequences were aligned using the ClustalW method. Phylogenetic analyses were performed using MEGA 5.0 using the neighbor joining method and tree topology was verified with 1000 bootstrap replicates (Tamura K, et al. 2011). HA gene sequences were deposited to Genbank under accession numbers JQ638655-JQ638665.

**Reverse Genetics.** In the context of molecular biology, “reverse genetics” is defined as the generation of virus possessing a genome derived from cloned cDNAs (for a review, see Neumann et al., *J. Gen. Viral.*, 83:2635; 2002). In the instant study, a triple reassortant internal gene cassette (TRIG) swine influenza virus (A/swine/North Carolina/3793/08(H1N1)) was used as the template to create a TRIG swine influenza virus reverse genetics system. All eight segments were PCR amplified, digested with BsmBI and ligated into a similarly digested pHW2000 as previously described (Hoffmann E, et al. 2001). Plasmids bearing insert were identified by restriction digest and sequenced to verify identity with A/swine/North Carolina/3793/08. The HA and NA genes (SEQ ID NOs: 7 & 11) from 10-0036-2 were also cloned into pHW2000 (Hoffmann et al 2000, PNAS 97(11):

6108-6113) and transfected along with plasmids bearing polymerase basic 2 (PB2), polymerase basic 1 (PB1), polymerase acid (PA), nucleoprotein (NP), matrix (M) and non-structural genes (NS) derived from A/swine/North Carolina/3793/08 to produce reverse genetics-derived 10-0036-2 (RG 10-0036-2, FIG. 1). Site directed mutagenesis was performed on the plasmid containing HA gene from 10-0036-2 to create mutants with an additional 1-5 N-linked glycosylation sites using the Quik Change II Site Directed Mutagenesis kit (Agilent Technologies) (Table 1, FIG. 2). Glycans are added to proteins at asparagine (N) residues located in the context of the N-linked glycosylation motif of N-X-S/T, where N is the amino acid asparagine, X is any amino acid and S/T is serine or threonine.

TABLE 1

Additional N-linked glycosylation sites engineered into the HA gene of RG10-0036-2 using site directed mutagenesis			
	Number additional N-linked glycosylation sites	Nucleotide Change	Amino Acid Change
20	1	G212A	S71N
25	2	G212A, G270T	S71N, K90N
30	3	G212A, G270T, CT517-518AC	S71N, K90N, L173T
35	4	G212A, G270T, CT517-518AC, C859A	S71N, K90N, L173T, P287T
40	5	G212A, G270T, CT517-518AC, C859A, A881C	S71N, K90N, L173T, P287T, K294T

Rescue of recombinant viruses was performed as previously described (Hoffmann E, et al. 2000). In brief, 293T and MDCK cells were co-cultured in Opti-MEM I containing 5% FBS in 6-well plates approximately 1×10<sup>6</sup> cells of each 293T and MDCK approximately 18 hours prior to transfection. One hundred nanograms of each of the eight plasmids were pooled in 100 µL of Opti-MEM I and combined with 100 µL Opti-MEM containing 3 µL Lipofectamine (Invitrogen) and incubated at room temperature 15 minutes before being diluted to 1 mL with Opti-MEM I and transferred to a single well of the 6-well plate. Plates were incubated at 37° C. with 5% CO<sub>2</sub> for 6 hours before the transfection mixture was replaced with Opti-MEM I. At 24 hours post transfection, 1.5 mL was transferred to a 6-well plate of confluent MDCK cells and 1.5 mL of DMEM containing 1 µg/mL of TPCK-treated trypsin was added. Viruses were harvested on day 5 post infection and their titers determined by the HA assay. The HA genes of rescued viruses were sequenced to verify the correct sequence.

**Results.** Genetic analysis of predicted N-linked glycosylation sites (N-X-S/T) found sites at N28, N40, N104, N142, N176, N303, N497 and N556 for virus 10-0036-2. Site directed mutagenesis was used to add an additional 1-5 N-linked glycosylation sites to the globular head portion of HA (Table 1). Following virus rescue from cell culture, mutant viruses were characterized by growth studies on ST cells. Growth studies were performed as attenuated viruses often demonstrate decreased growth rates and titers in vitro. Mutant viruses with 4 or 5 additional N-linked glycosylation sites showed such growth defects, suggesting that additional glycosylation attenuated the viruses. The attenuated viruses were next evaluated in swine to evaluate their virulence in vivo and characterize the immune response against these mutant viruses.

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## Example 2

## Efficacy of Attenuated Swine Influenza Vaccines in Pigs

**Materials & Methods.** Sixty 3-week old high health pigs (confirmed SIV seronegative by IDEXX FlockChek ELISA) were separated into 4 groups of 15 in separate rooms. On day 0 (d0), pigs were inoculated intranasally with 2 mL of 6.0 TCID<sub>50</sub>/mL virus. Group 1 was mock infected with cell culture media (DMEM). Group 2 received 10-0036-2 n+5. Group 3 received 10-0036-2 n+4 (a mutant with 4 additional glycosylation sites; similar to n+5 but lacking the mutation A881C [K294I]). Group 4 received reverse genetics created 10-0036-2 parent (no mutations). Pigs were swabbed (nasal) at day 0 and samples were run by QPCR for SIV detection to verify no active infection. Results are summarized in Table 2. For Tables 2 and 3, groups with different letters have statistically different means (P<0.05). For example, "A" is statistically different from "B", and "BC" is statistically different from "A" but not "B" or "C".

TABLE 2

Vaccination (H1N2)	Vaccination study				
	Nasal Swab, Day 3 (TCID <sub>50</sub> /mL)	Nasal Swab, Day 5 (TCID <sub>50</sub> / mL)	Lung Titer, Day 5 (TCID <sub>50</sub> / mL)	Lung Score	IHC Score
Neg. Con	0.0 A	0.0 A	0.0 A	0.0 A	0.0 A
n + 4 mutant	3.1 B	2.8 B	0.9 A	0.0 A	0.0 A
n + 5 mutant	2.6 C	2.7 B	0.4 A	0.2 A	0.2 A
parent	4.1 D	3.2 C	5.4 B	1.5 B	2.0 B

Nasal swabs collected on days 1, 3 and 5. Five pigs from each group were euthanized at day 5 and lung samples were collected. Swabs from day 1 were analyzed by QPCR. Swabs from days 3 and 5 as well as lung samples were titrated for SIV. Lungs were sent to a University Diagnostic lab for histopathological analysis and IHC. On day 21 pigs were revaccinated as above. These results demonstrate the mutants containing 4 or 5 additionally N-linked glycosylation sites are attenuated and avirulent in swine, in agreement with the in vitro growth studies. Nasal swab titrations indicated that the

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virus was capable of replicating in vivo, however, absence of virus in lungs and lack of lung damage suggest replication limited to the upper respiratory tract. Mutations in other influenza genes that confer temperature sensitivity have been shown to limit infection to the upper respiratory tract and are the basis of the human live attenuated influenza vaccine Flu-Mist™.

On day 31, pigs were challenged with a field isolate 12-1110-1 (H3N2). The results of the challenge study are summarized in Table 3. For the challenge (performed comparably to Richt et al 2006, J. Virology 80(22):11009-11018) 2mL of 4.6 TCID<sub>50</sub>/mL was delivered intranasally. Blood and nasal swabs were collected on day 31 prior to challenge. Nasal swabs were collected on days 0 and 1, and analyzed by QPCR. Nasal swabs were collected on days 3 and 5 and SIV titer determined by titration. All pigs were euthanized on day 5 and lung samples analyzed by titration. Lung samples analyzed as above.

TABLE 3

H3N2 Challenge	Challenge study				
	Nasal Swab, Day 3 (TCID <sub>50</sub> /mL)	Nasal Swab, Day 5 (TCID <sub>50</sub> / mL)	Lung Titer, Day 5 (TCID <sub>50</sub> / mL)	Lung Score	IHC Score
Neg. Con	5.4 A	5.8 A	3.0 A	1.6 A	1.5 A
n + 4 mutant	2.3 B	0.7 B	0.0 B	0.0 B	0.0 B
n + 5 mutant	2.9 BC	1.8 C	0.1 B	0.0 B	0.0 B
parent	0.5 C	0.9 BC	0.0 B	0.0 B	0.0 B

These results demonstrate that pigs vaccinated with mutants n+4 or n+5 were protected from disease as evident by lack of virus in lungs by titration and IHC, as well as no evidence of lung lesions. Naïve (negative control) pigs were readily infected with the H3N2 challenge virus and demonstrated classical influenza disease. Pigs previously infected with the parent virus 10-0036-2 were also protected from the H3N2 challenge.

Having thus described in detail preferred embodiments of the present invention, it is to be understood that the invention defined by the above paragraphs is not to be limited to particular details set forth in the above description as many apparent variations thereof are possible without departing from the spirit or scope of the present invention.

## SEQUENCE LISTING

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<212> TYPE: DNA
<213> ORGANISM: Influenza A virus

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&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Influenza A virus

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35	40	45

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 305 310 315 320  
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 Lys Ile Arg Val His Glu Gly Tyr Glu Glu Phe Thr Met Val Gly Arg  
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 <211> LENGTH: 2271  
 <212> TYPE: DNA  
 <213> ORGANISM: Influenza A virus

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&lt;211&gt; LENGTH: 757

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 4

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Gly Thr Gly Thr Gly Tyr Thr Met Asp Thr Val Asn Arg Thr His Gln		
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Glu Ser His Pro Gly Ile Phe Glu Asn Ser Cys Leu Glu Thr Met Glu  
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Ile Val Gln Gln Thr Arg Val Asp Lys Leu Thr Gln Gly Arg Gln Thr  
115 120 125

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Gly Arg Leu Ile Asp Phe Leu Lys Asp Val Met Glu Ser Met Asp Lys  
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Glu Glu Ile Glu Ile Thr Thr His Phe Gln Arg Lys Arg Arg Val Arg  
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210 215 220

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<210> SEQ ID NO 5  
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 <212> TYPE: DNA  
 <213> ORGANISM: Influenza A virus

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ttgaagacaa caccacgccc tctaaaatg cctgatggc ctccctgctc tcagggctca 840  
aagttcttgc tgatggatgc tctgaaacta agtattgaag acccgagtca tgaaggagaa 900  
ggaataccac tatatgtatgc aatcaagtgc atgaagacat tttttggctg gaaagaaccc 960  
aacataatca aaccacatga gaaaggcata aaccccaatt acctactggc ttgaaagcag 1020  
gtgcttagcag agctccaaga cattgaaaat gaagagaaga tcccaaagac aaagaacatg 1080  
aggagaacaa gccaattgaa gtgggcaactc ggtgagaata tggcaccaga gaaagttagat 1140  
tttgcgtact gcaaagatgt tggtgatctt aaacagtatg acagcgcacga gccagagccc 1200  
agatctctag caagttgggt cccaaatgaa ttcaacaagg catgtgaatt gaccgattca 1260  
agctggatag aacttgcgtga gataggagaa gatattgcac cgattgaaca catgcgaatg 1320  
atgaggagga actatttac agcagaagtgc tcccattgtt gggctacggc atacataatg 1380  
aaggaggatgt acataaacac ggcttgctt aatgcacattt gtgcagccat ggtactttt 1440  
cagctgatcc caatgataag caaatgcagg accaaagaag gaagacgaaa aacaaatctg 1500  
tatgggttca ttataaaagg aaggtcccat ctgaggaatg atactgacgt ggtgaacttt 1560  
gtaaagcatgg agttctccct caccgacccg agactggagc cacacaaatg gaaaaatac 1620  
tgtgttcttgg aaataggaga catgtctcg aggactgoga taggcacatg gtcggggccc 1680  
atgttcttat atgtgagaac caatggaaacc tccaagatca agatgaaatg gggcatggaa 1740  
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tctgtaaaag agaaaagacat gaccaaggaa tttttgaaa acaaatcgga aacatggcca 1860  
atgggagaat cacccaaagg agtggaggaa ggctctattt gggaaatgtg caggacctta 1920  
ctggcaaaat ctgttattcaa cagtctatac gctctccac aacttgaggg atttcggt 1980  
gaatcgagaa agttgttctt cattgttgc gcaacttaggg acaacctggaa acctggaaacc 2040  
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ttgtttaatg catcttgggtt caactccctc ctcacacatg cactgaaa 2148

<210> SEQ ID NO 6  
<211> LENGTH: 716  
<212> TYPE: PRT  
<213> ORGANISM: Influenza A virus

<400> SEQUENCE: 6

1                   5                   10                   15  
 Ala Glu Lys Ala Met Lys Glu Tyr Gly Glu Asp Pro Lys Ile Glu Thr

Asn Lys Phe Ala Ala Ile Cys Thr His Leu Glu Val Cys Phe Met Tyr

Ser Asp Phe His Phe Ile Asp Glu Arg Gly Glu Ser Ile Ile Val Glu

Ser Gly Asp Pro Asn Ala Leu Leu Lys His Arg Phe Glu Ile Ile Glu  
65 70 75 80

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Gly Arg Asp Arg Thr Met Ala Trp Thr Val Val Asn Ser Ile Cys Asn  
85 90 95

Thr Thr Gly Val Glu Lys Pro Lys Phe Leu Pro Asp Leu Tyr Asp Tyr  
100 105 110

Lys Glu Asn Arg Phe Ile Glu Ile Gly Val Thr Arg Arg Glu Val His  
115 120 125

Ile Tyr Tyr Leu Glu Lys Ala Asn Lys Ile Lys Ser Glu Lys Thr His  
130 135 140

Ile His Ile Phe Ser Phe Thr Gly Glu Glu Met Ala Thr Lys Ala Asp  
145 150 155 160

Tyr Thr Leu Asp Glu Glu Ser Arg Ala Arg Ile Lys Thr Arg Leu Phe  
165 170 175

Thr Ile Arg Gln Glu Met Ala Ser Arg Gly Leu Trp Asp Ser Phe Arg  
180 185 190

Gln Ser Glu Arg Gly Glu Glu Thr Ile Glu Glu Arg Phe Glu Ile Thr  
195 200 205

Gly Thr Met Arg Arg Leu Ala Asp Gln Ser Leu Pro Pro Asn Phe Ser  
210 215 220

Ser Leu Glu Asn Phe Arg Ala Tyr Val Asp Gly Phe Glu Pro Asn Gly  
225 230 235 240

Cys Ile Glu Gly Lys Leu Ser Gln Met Ser Lys Glu Val Ser Ala Gln  
245 250 255

Ile Glu Pro Phe Leu Lys Thr Thr Pro Arg Pro Leu Lys Leu Pro Asp  
260 265 270

Gly Pro Pro Cys Ser Gln Arg Ser Lys Phe Leu Leu Met Asp Ala Leu  
275 280 285

Lys Leu Ser Ile Glu Asp Pro Ser His Glu Gly Glu Gly Ile Pro Leu  
290 295 300

Tyr Asp Ala Ile Lys Cys Met Lys Thr Phe Phe Gly Trp Lys Glu Pro  
305 310 315 320

Asn Ile Ile Lys Pro His Glu Lys Gly Ile Asn Pro Asn Tyr Leu Leu  
325 330 335

Ala Trp Lys Gln Val Leu Ala Glu Leu Gln Asp Ile Glu Asn Glu Glu  
340 345 350

Lys Ile Pro Lys Thr Lys Asn Met Arg Arg Thr Ser Gln Leu Lys Trp  
355 360 365

Ala Leu Gly Glu Asn Met Ala Pro Glu Lys Val Asp Phe Asp Asp Cys  
370 375 380

Lys Asp Val Gly Asp Leu Lys Gln Tyr Asp Ser Asp Glu Pro Glu Pro  
385 390 395 400

Arg Ser Leu Ala Ser Trp Val Gln Asn Glu Phe Asn Lys Ala Cys Glu  
405 410 415

Leu Thr Asp Ser Ser Trp Ile Glu Leu Asp Glu Ile Gly Glu Asp Ile  
420 425 430

Ala Pro Ile Glu His Ile Ala Ser Met Arg Arg Asn Tyr Phe Thr Ala  
435 440 445

Glu Val Ser His Cys Arg Ala Thr Glu Tyr Ile Met Lys Gly Val Tyr  
450 455 460

Ile Asn Thr Ala Leu Leu Asn Ala Ser Cys Ala Ala Met Asp Asp Phe  
465 470 475 480

Gln Leu Ile Pro Met Ile Ser Lys Cys Arg Thr Lys Glu Gly Arg Arg  
485 490 495

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Lys Thr Asn Leu Tyr Gly Phe Ile Ile Lys Gly Arg Ser His Leu Arg  
500 505 510

Asn Asp Thr Asp Val Val Asn Phe Val Ser Met Glu Phe Ser Leu Thr  
515 520 525

Asp Pro Arg Leu Glu Pro His Lys Trp Glu Lys Tyr Cys Val Leu Glu  
530 535 540

Ile Gly Asp Met Leu Leu Arg Thr Ala Ile Gly Gln Val Ser Arg Pro  
545 550 555 560

Met Phe Leu Tyr Val Arg Thr Asn Gly Thr Ser Lys Ile Lys Met Lys  
565 570 575

Trp Gly Met Glu Met Arg Arg Cys Leu Leu Gln Ser Leu Gln Gln Ile  
580 585 590

Glu Ser Met Ile Glu Ala Glu Ser Ser Val Lys Glu Lys Asp Met Thr  
595 600 605

Lys Glu Phe Phe Glu Asn Lys Ser Glu Thr Trp Pro Ile Gly Glu Ser  
610 615 620

Pro Lys Gly Val Glu Glu Gly Ser Ile Gly Lys Val Cys Arg Thr Leu  
625 630 635 640

Leu Ala Lys Ser Val Phe Asn Ser Leu Tyr Ala Ser Pro Gln Leu Glu  
645 650 655

Gly Phe Ser Ala Glu Ser Arg Lys Leu Leu Ile Val Gln Ala Leu  
660 665 670

Arg Asp Asn Leu Glu Pro Gly Thr Phe Asp Leu Gly Leu Tyr Glu  
675 680 685

Ala Ile Glu Glu Cys Leu Ile Asn Asp Pro Trp Val Leu Leu Asn Ala  
690 695 700

Ser Trp Phe Asn Ser Phe Leu Thr His Ala Leu Lys  
705 710 715

<210> SEQ\_ID NO 7  
<211> LENGTH: 1692  
<212> TYPE: DNA  
<213> ORGANISM: Influenza A virus

<400> SEQUENCE: 7

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atgaaggcaa tactagtagt cctgctatac acatttacaa ccgcaaatgc cgacacatta      60
tgcatacggtt atcatgcgaa caattcaact gacaccgtag acacagtgt agaaaaaaaaat    120
gtaacagtaa cacactctgt caacacctcta gaaaacaggc ataatggaa actatgtaaa    180
ctaagagggg tagctccatt gcattttgggt aaatgtaaaca ttgctggctg gctcctggga    240
aatccagagt gtgagtcaat ctccaaagca agtcgtatgg cctacattgt ggaaacatct    300
aattcagaca atggcacgtg ttacccagga gatttcatca attatgagga gctaagagag    360
cagttgagct cagtgtcatc atttggaaaga tttgagatat tccccatgac aagttcatgg    420
cccaatcatg acacgaacag aggtgtgacg gcagcatgtc ctcacgtgg gacaaatagc    480
ttctacaaaa atttaatatg gctggtcaaa aaaggaaatt catacccaa gatcaacaaa    540
tcctacatta acaacaaaga gaaagaagtt ctcgtgtat gggccataca tcatccacct    600
accaatgccg accaacaaag cctctaccaa aatgcagatg cctatgttt tgtgggtca    660
tcaagataca gcaggaagtt cgagccagaa atagcaacaa gacccaaggt gagagaccaa    720
gcagggagaa tgaactatta ctggacattg gtagagcctg gagacaagat aacattcgaa    780
gcaactggaa atctagtggt accgagatat gccttcgcat tgaaaagaaa ttctggatct    840
ggttattatca tttcagatac atcagtccac gattgtgata cgacttgtca gacacccaaat   900

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gggtctataa acaccagcct cccatttcaa aatatacatc cagtacaat tggagaatgt	960
ccaaaatatg taaaaagtac taaactgaga atggccacag gattaaggaa tatcccgct	1020
attcaatcta gaggcctgtt tggggccatt gctggctta ttgaaggggg ctggcacgga	1080
atgatagacg gatggtacgg ttaccaccaat caaaatgagc agggatcagg atatgcagcc	1140
gacctgaaaa gcacacagaa tgccattgac gggatcacta acaaggtaaa ttctgttatt	1200
gaaaagatga acacacaatt cacagcagta ggtaaagagt tcagccactt ggaaagaaga	1260
atagagaatt taaataaaaaa ggttgatgat ggtttctag atatttggac ttacaatgcc	1320
gaactgttgg ttctgttggaa gaatgaaaga actttggatt accacgattc aaatgtgaaa	1380
aacttatatg aaaaagtaag aagccaacta aaaaacaatg ccaaagaaat tggaaatggc	1440
tgotttgaat ttaccaccaa atgtgatgac acgtgtatgg aaagcgtcaa aaatgggact	1500
tatgattacc caaaaactc agaggaagca aaactaaaca gagagggaaat agatggggta	1560
aagttggaaat caacaagggt ttaccaaatt ttggcgatct attcaacgggt cgccgatc	1620
ttggtactgg tagtctccct gggggcaatc agtttctgga tgtgctctaa tgggtcgcta	1680
cagtqcaqaa ta	1692

<210> SEQ ID NO 8  
<211> LENGTH: 564  
<212> TYPE: PRT  
<213> ORGANISM: Influenza A virus

<400> SEQUENCE: 8

Met	Lys	Ala	Ile	Leu	Val	Val	Leu	Leu	Tyr	Thr	Phe	Thr	Thr	Ala	Asn
1									10						15
Ala	Asp	Thr	Leu	Cys	Ile	Gly	Tyr	His	Ala	Asn	Asn	Ser	Thr	Asp	Thr
			20					25					30		
Val	Asp	Thr	Val	Leu	Glu	Lys	Asn	Val	Thr	Val	Thr	His	Ser	Val	Asn
			35			40						45			
Leu	Leu	Glu	Asn	Arg	His	Asn	Gly	Lys	Leu	Cys	Lys	Leu	Arg	Gly	Val
			50			55						60			
Ala	Pro	Leu	His	Leu	Gly	Lys	Cys	Asn	Ile	Ala	Gly	Trp	Leu	Leu	Gly
	65				70					75					80
Asn	Pro	Glu	Cys	Glu	Ser	Ile	Ser	Lys	Ala	Ser	Ser	Trp	Ser	Tyr	Ile
		85						90					95		
Val	Glu	Thr	Ser	Asn	Ser	Asp	Asn	Gly	Thr	Cys	Tyr	Pro	Gly	Asp	Phe
		100						105					110		
Ile	Asn	Tyr	Glu	Glu	Leu	Arg	Glu	Gln	Leu	Ser	Ser	Val	Ser	Ser	Phe
		115				120						125			
Glu	Arg	Phe	Glu	Ile	Phe	Pro	Met	Thr	Ser	Ser	Trp	Pro	Asn	His	Asp
	130					135						140			
Thr	Asn	Arg	Gly	Val	Thr	Ala	Ala	Cys	Pro	His	Ala	Gly	Thr	Asn	Ser
	145				150					155					160
Phe	Tyr	Lys	Asn	Leu	Ile	Trp	Leu	Val	Lys	Lys	Gly	Asn	Ser	Tyr	Pro
			165						170				175		
Lys	Ile	Asn	Lys	Ser	Tyr	Ile	Asn	Asn	Lys	Glu	Lys	Glu	Val	Leu	Val
			180						185				190		
Leu	Trp	Ala	Ile	His	His	Pro	Pro	Thr	Asn	Ala	Asp	Gln	Gln	Ser	Leu
		195				200						205			
Tyr	Gln	Asn	Ala	Asp	Ala	Tyr	Val	Phe	Val	Gly	Ser	Ser	Arg	Tyr	Ser
	210					215						220			

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Arg Lys Phe Glu Pro Glu Ile Ala Thr Arg Pro Lys Val Arg Asp Gln  
 225 230 235 240  
 Ala Gly Arg Met Asn Tyr Tyr Trp Thr Leu Val Glu Pro Gly Asp Lys  
 245 250 255  
 Ile Thr Phe Glu Ala Thr Gly Asn Leu Val Val Pro Arg Tyr Ala Phe  
 260 265 270  
 Ala Leu Lys Arg Asn Ser Gly Ser Gly Ile Ile Ser Asp Thr Ser  
 275 280 285  
 Val His Asp Cys Asp Thr Thr Cys Gln Thr Pro Asn Gly Ala Ile Asn  
 290 295 300  
 Thr Ser Leu Pro Phe Gln Asn Ile His Pro Val Thr Ile Gly Glu Cys  
 305 310 315 320  
 Pro Lys Tyr Val Lys Ser Thr Lys Leu Arg Met Ala Thr Gly Leu Arg  
 325 330 335  
 Asn Ile Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly  
 340 345 350  
 Phe Ile Glu Gly Gly Trp Thr Gly Met Ile Asp Gly Trp Tyr Gly Tyr  
 355 360 365  
 His His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Leu Lys Ser  
 370 375 380  
 Thr Gln Asn Ala Ile Asp Gly Ile Thr Asn Lys Val Asn Ser Val Ile  
 385 390 395 400  
 Glu Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Ser His  
 405 410 415  
 Leu Glu Arg Arg Ile Glu Asn Leu Asn Lys Lys Val Asp Asp Gly Phe  
 420 425 430  
 Leu Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn  
 435 440 445  
 Glu Arg Thr Leu Asp Tyr His Asp Ser Asn Val Lys Asn Leu Tyr Glu  
 450 455 460  
 Lys Val Arg Ser Gln Leu Lys Asn Asn Ala Lys Glu Ile Gly Asn Gly  
 465 470 475 480  
 Cys Phe Glu Phe Tyr His Lys Cys Asp Asp Thr Cys Met Glu Ser Val  
 485 490 495  
 Lys Asn Gly Thr Tyr Asp Tyr Pro Lys Tyr Ser Glu Glu Ala Lys Leu  
 500 505 510  
 Asn Arg Glu Glu Ile Asp Gly Val Lys Leu Glu Ser Thr Arg Val Tyr  
 515 520 525  
 Gln Ile Leu Ala Ile Tyr Ser Thr Val Ala Ser Ser Leu Val Leu Val  
 530 535 540  
 Val Ser Leu Gly Ala Ile Ser Phe Trp Met Cys Ser Asn Gly Ser Leu  
 545 550 555 560  
 Gln Cys Arg Ile

<210> SEQ\_ID NO 9  
 <211> LENGTH: 1512  
 <212> TYPE: DNA  
 <213> ORGANISM: Influenza A virus

<400> SEQUENCE: 9

atgagtgtaca tcgaagccat ggcgtctcaa ggcaccaaac gatcatatga acaaatggag	60
actgggtggtg aacgccagga tgccacagaa atcagagcat ctgtcgaaag aatgatttgt	120
ggaatcggga aattctacat ccaaattgtgc actgaactca aactcagtga ctatgaggaa	180

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cgactaatcc	aaaatagcat	aacaatagag	agaatggtgc	tctctgttt	tgatgagaga	240
agaaataaat	acctagaaga	gcatcccagt	gctgggaaag	accctaagaa	aactggagga	300
ccccatatata	gaagagtaga	cggaaagtgg	atgagagaac	tcattttta	tgacaaagaa	360
gaaaataagga	gagttggcg	ccaggcaaac	aatggtgatg	atgcaacagc	tggcttact	420
catatcatga	tttggcatc	caatctgaat	gatgccacgt	accagagaac	aagagcactt	480
gttcgcaccg	aatggatcc	cagaatgtgc	tctctaattgc	aaggttcaac	acttccaga	540
aggtctggag	cagcaggtgc	tgcagtgaaa	ggagttggaa	caataacaat	ggaattaatc	600
agaatgatca	aacggggat	caatgaccga	aatttctgga	gaggtgaaaa	tggaagaagg	660
acaaggattg	catatgaaag	aatgtgcaat	attctcaaag	gaaaatttca	gacagctgcc	720
caaaggcga	tgtggatca	agtggagaa	agtcggAAC	caggAACG	tgagattgaa	780
gatctcattt	tcctggcacg	gtcagcacctt	atcctaagggg	gatcagttgc	acataagtct	840
tgcctgcctg	cttgcgtgta	tgggcttgca	gtggcaagtg	ggcatgactt	tgaaaggaa	900
gggttattcgc	tggtcggat	agaccattt	aaattactcc	agaacagtca	agtgttcac	960
ctggtaagac	caaataaaaa	cccagctcac	aagagtcaat	tagtgtggat	ggcatgcccac	1020
tctgctgcat	ttgaggatct	aagggtctca	agtttcataa	gagggaagaa	agtgttcca	1080
aggggaaagc	tttccacaag	aggggttcag	attgcttcaa	atgagaatgt	ggaagccatg	1140
gattccaata	ccttagagct	gagaagcaga	tactgggcta	taaggaccag	aagtggagga	1200
aatactaatac	aacagaaagc	atccgcaggc	cagatcagtg	tgcaacctac	attctcagtg	1260
caacggaaatc	tccctttga	aagagcaacc	gttatggcag	ctttcagccg	aaacaatgaa	1320
ggacggacat	ccgatatgcf	gacagaaatt	ataaggatga	tggaaaatgc	aaaaccagaa	1380
gatttgcctt	cccaggggcg	gggagtcttc	gagctctcg	acgaaaagc	aacgagcccg	1440
atcgtgcctt	ccttgacat	gagtaatgaa	gggtcttatt	tcttcggaga	caatgcagag	1500
gagtatgaca	gt					1512

&lt;210&gt; SEQ\_ID NO 10

&lt;211&gt; LENGTH: 504

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 10

Met	Ser	Asp	Ile	Glu	Ala	Met	Ala	Ser	Gln	Gly	Thr	Lys	Arg	Ser	Tyr
1						5			10			15			

Glu	Gln	Met	Glu	Thr	Gly	Gly	Glu	Arg	Gln	Asp	Ala	Thr	Glu	Ile	Arg
		20					25					30			

Ala	Ser	Val	Gly	Arg	Met	Ile	Gly	Ile	Gly	Lys	Phe	Tyr	Ile	Gln	
					35			40				45			

Met	Cys	Thr	Glu	Leu	Lys	Leu	Ser	Asp	Tyr	Glu	Gly	Arg	Leu	Ile	Gln
					50			55			60				

Asn	Ser	Ile	Thr	Ile	Glu	Arg	Met	Val	Leu	Ser	Ala	Phe	Asp	Glu	Arg
65					70			75			80				

Arg	Asn	Lys	Tyr	Leu	Glu	Glu	His	Pro	Ser	Ala	Gly	Lys	Asp	Pro	Lys
							85		90			95			

Lys	Thr	Gly	Gly	Pro	Ile	Tyr	Arg	Arg	Val	Asp	Gly	Lys	Trp	Met	Arg
					100			105			110				

Glu	Leu	Ile	Leu	Tyr	Asp	Lys	Glu	Ile	Arg	Arg	Val	Trp	Arg	Gln	
					115			120			125				

Ala Asn Asn Gly Asp Asp Ala Thr Ala Gly Leu Thr His Ile Met Ile

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130	135	140
Trp His Ser Asn Leu Asn Asp Ala Thr Tyr Gln Arg Thr Arg Ala Leu		
145	150	155
160		
Val Arg Thr Gly Met Asp Pro Arg Met Cys Ser Leu Met Gln Gly Ser		
165	170	175
Thr Leu Pro Arg Arg Ser Gly Ala Ala Gly Ala Ala Val Lys Gly Val		
180	185	190
Gly Thr Ile Thr Met Glu Leu Ile Arg Met Ile Lys Arg Gly Ile Asn		
195	200	205
Asp Arg Asn Phe Trp Arg Gly Glu Asn Gly Arg Arg Thr Arg Ile Ala		
210	215	220
Tyr Glu Arg Met Cys Asn Ile Leu Lys Gly Lys Phe Gln Thr Ala Ala		
225	230	235
240		
Gln Arg Ala Met Met Asp Gln Val Arg Glu Ser Arg Asn Pro Gly Asn		
245	250	255
Ala Glu Ile Glu Asp Leu Ile Phe Leu Ala Arg Ser Ala Leu Ile Leu		
260	265	270
Arg Gly Ser Val Ala His Lys Ser Cys Leu Pro Ala Cys Val Tyr Gly		
275	280	285
Leu Ala Val Ala Ser Gly His Asp Phe Glu Arg Glu Gly Tyr Ser Leu		
290	295	300
Val Gly Ile Asp Pro Phe Lys Leu Leu Gln Asn Ser Gln Val Phe Ser		
305	310	315
320		
Leu Val Arg Pro Asn Glu Asn Pro Ala His Lys Ser Gln Leu Val Trp		
325	330	335
Met Ala Cys His Ser Ala Ala Phe Glu Asp Leu Arg Val Ser Ser Phe		
340	345	350
Ile Arg Gly Lys Lys Val Ile Pro Arg Gly Lys Leu Ser Thr Arg Gly		
355	360	365
Val Gln Ile Ala Ser Asn Glu Asn Val Glu Ala Met Asp Ser Asn Thr		
370	375	380
Leu Glu Leu Arg Ser Arg Tyr Trp Ala Ile Arg Thr Arg Ser Gly Gly		
385	390	395
400		
Asn Thr Asn Gln Gln Lys Ala Ser Ala Gly Gln Ile Ser Val Gln Pro		
405	410	415
Thr Phe Ser Val Gln Arg Asn Leu Pro Phe Glu Arg Ala Thr Val Met		
420	425	430
Ala Ala Phe Ser Gly Asn Asn Glu Gly Arg Thr Ser Asp Met Arg Thr		
435	440	445
Glu Ile Ile Arg Met Met Glu Asn Ala Lys Pro Glu Asp Leu Ser Phe		
450	455	460
Gln Gly Arg Gly Val Phe Glu Leu Ser Asp Glu Lys Ala Thr Ser Pro		
465	470	475
480		
Ile Val Pro Ser Phe Asp Met Ser Asn Glu Gly Ser Tyr Phe Phe Gly		
485	490	495
Asp Asn Ala Glu Glu Tyr Asp Ser		
500		

<210> SEQ ID NO 11  
<211> LENGTH: 1407  
<212> TYPE: DNA  
<213> ORGANISM: Influenza A virus  
<400> SEQUENCE: 11

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agtctattgt tacagatagg aaacatggtt tcgttatgga tcagccatc aattcagact	120
gaagggaaaa atcatactga gatgtgcaat caaaatgtca ttacatatgt aaataacaca	180
tgggtgaacc gaacttatgt aaacattagc aataccaaa ttgttaatgt acaggacgtg	240
gttcagtaa tattaacgg caattccctc ctctgccaa taagtgggtg ggctatatac	300
agcaaagaca atagcataag gattgggtct aaagggaca ttttgcata aagagaacca	360
ttcatttcat gctctcaattt ggaatgcaga acttttttc tgacccaagg cgcttgctg	420
aatgacaaggc attctaattgg aaccgtcaag gacaggagtc cctatagaac tttaatgagc	480
tgtccccatcg gtgaagctcc atctccatat aactcaaggc tcgaatcagt tgcttggca	540
gcaaggtgcattt gccatgtgg gatggatgg ctaacaatcg gaatctccgg tccagataat	600
ggagcagtag ctgtttaaa atacaacggc ataataacag atacaataaa aagttggaga	660
aacaaaatataa taagaacaca agagtcagaa tgtgtttgtt tgaacgggtc ttgtttact	720
gtattaaactg atggcccaag caatggcaag gcacgttaca aaatattcaa gatggaaaaa	780
ggaaaaataaa ttaaatcaat tgagctggat gcacccaaattt accactatga ggaatgctcc	840
tgttatccctg atgcaggcaaa agtaatgtgt gtttgcagag acaactggca tgcctcgaaac	900
cggccatggg tctcttcga tcagaatctt aattatcaaa tagggtacat atgcagtgg	960
gttttcgggtg ataaccgcgc ttctaatgtt gaaaggccc atttgtggcc accatattct	1020
aatggagcaaa atggagtgaa aggattctca tataaatatg gtaatgggtt ttggatagga	1080
aggactaaaa gatcaactc cagaagtggaa ttgaaatgtt ttggatggcc aatgggtgg	1140
actggaaactg atagtagttt ctctatgtt caggatatta tagcattaa tgattggca	1200
ggatacagtg gaagttttgtt ccaacatctt gaattaaacag gaatgaatgg cataaggccc	1260
tgtttctggg tagaattaaat cagaggcaaa ccaaggaaa acaccatctg ggctagcgaa	1320
agcagcatctt ctttctgtgg taaaatgtt gaaaccggcaaa gctggcatg gccagacggaa	1380
gtgtatctgc cattcaccat tgacaag	1407

&lt;210&gt; SEQ ID NO 12

&lt;211&gt; LENGTH: 469

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 12

Met	Asn	Thr	Asn	Gln	Arg	Ile	Ile	Thr	Ile	Gly	Thr	Val	Cys	Leu	Ile
						5		10					15		

Val	Gly	Ile	Ile	Ser	Leu	Leu	Leu	Gln	Ile	Gly	Asn	Met	Val	Ser	Leu
						20		25				30			

Trp	Ile	Ser	His	Ser	Ile	Gln	Thr	Glu	Gly	Lys	Asn	His	Thr	Glu	Met
						35		40			45				

Cys	Asn	Gln	Asn	Val	Ile	Thr	Tyr	Val	Asn	Asn	Thr	Trp	Val	Asn	Arg
						50		55			60				

Thr	Tyr	Val	Asn	Ile	Ser	Asn	Thr	Lys	Ile	Val	Asn	Val	Gln	Asp	Val
						65		70			75		80		

Val	Ser	Val	Ile	Leu	Thr	Gly	Asn	Ser	Ser	Leu	Cys	Pro	Ile	Ser	Gly
						85		90			95				

Trp	Ala	Ile	Tyr	Ser	Lys	Asp	Asn	Ser	Ile	Arg	Ile	Gly	Ser	Lys	Gly
						100		105			110				

Asp	Ile	Phe	Val	Ile	Arg	Glu	Pro	Phe	Ile	Ser	Cys	Ser	His	Leu	Glu
						115		120			125				

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Cys Arg Thr Phe Phe Leu Thr Gln Gly Ala Leu Leu Asn Asp Lys His  
130 135 140

Ser Asn Gly Thr Val Lys Asp Arg Ser Pro Tyr Arg Thr Leu Met Ser  
145 150 155 160

Cys Pro Ile Gly Glu Ala Pro Ser Pro Tyr Asn Ser Arg Phe Glu Ser  
165 170 175

Val Ala Trp Ser Ala Ser Ala Cys His Asp Gly Met Gly Trp Leu Thr  
180 185 190

Ile Gly Ile Ser Gly Pro Asp Asn Gly Ala Val Ala Val Leu Lys Tyr  
195 200 205

Asn Gly Ile Ile Thr Asp Thr Ile Lys Ser Trp Arg Asn Lys Ile Leu  
210 215 220

Arg Thr Gln Glu Ser Glu Cys Val Cys Met Asn Gly Ser Cys Phe Thr  
225 230 235 240

Val Leu Thr Asp Gly Pro Ser Asn Gly Gln Ala Ser Tyr Lys Ile Phe  
245 250 255

Lys Met Glu Lys Gly Lys Ile Ile Lys Ser Ile Glu Leu Asp Ala Pro  
260 265 270

Asn Tyr His Tyr Glu Glu Cys Ser Cys Tyr Pro Asp Ala Gly Lys Val  
275 280 285

Met Cys Val Cys Arg Asp Asn Trp His Ala Ser Asn Arg Pro Trp Val  
290 295 300

Ser Phe Asp Gln Asn Leu Asn Tyr Gln Ile Gly Tyr Ile Cys Ser Gly  
305 310 315 320

Val Phe Gly Asp Asn Pro Arg Ser Asn Asp Gly Lys Gly Asn Cys Gly  
325 330 335

Pro Val His Ser Asn Gly Ala Asn Gly Val Lys Gly Phe Ser Tyr Lys  
340 345 350

Tyr Gly Asn Gly Val Trp Ile Gly Arg Thr Lys Ser Ile Asn Ser Arg  
355 360 365

Ser Gly Phe Glu Met Ile Trp Asp Pro Asn Gly Trp Thr Gly Thr Asp  
370 375 380

Ser Ser Phe Ser Met Lys Gln Asp Ile Ile Ala Leu Thr Asp Trp Ser  
385 390 395 400

Gly Tyr Ser Gly Ser Phe Val Gln His Pro Glu Leu Thr Gly Met Asn  
405 410 415

Cys Ile Arg Pro Cys Phe Trp Val Glu Leu Ile Arg Gly Gln Pro Lys  
420 425 430

Glu Asn Thr Ile Trp Ala Ser Gly Ser Ser Ile Ser Phe Cys Gly Val  
435 440 445

Asn Gly Glu Thr Ala Ser Trp Ser Trp Pro Asp Gly Ala Asp Leu Pro  
450 455 460

Phe Thr Ile Asp Lys  
465

<210> SEQ ID NO 13  
<211> LENGTH: 756  
<212> TYPE: DNA  
<213> ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 13

atgagtccttc taaccgaggt cggaaacgtat gttctctcta tcgtcccgta agggccccctc 60  
aaagccgaga tagcacagag gctcgaagac gtttttgcaag ggaaaaaacac cgatctttag 120

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gctctcatgg aatggctaaa gacaagacca atcctgtcac ctctgactaa agggattta	180
gggttttgttgcac tcacgctcac cgtgccagt gagcgaggac tgcagegtac acgtttgtc	240
cagaatgccc tcaatggaa tggtgaccca aacaacatgg acaaggcgtt aaaaactgtac	300
aggaaactaa aaaggaaat aacattccat ggggccaagg aagttagcgct cagttactct	360
gtctggcac ttggcagttt catggccctc atatacaaca gaatggggac tgtcgcccact	420
gagggtggcat ttggctggat atgcgcacc tggtaacaaa ttgctgatc tcagcatcga	480
tctcatagac aaatggtgac aacaaccaat ccactaatca ggcacgagaa cagaatggta	540
atagccagca caacagctaa ggccatggaa caaatggctg gatcaagtga acaaggcagca	600
gaggctatgg aggttgccag tcaggctaga caaatggtac aggcaatgag aacaattggg	660
actcaccctt gttccagcac ttggctaaaa gatgatctt ttgaaaattt acaggcctat	720
cagaaacgga tgggagtgca aatgcaacga ttcaag	756

&lt;210&gt; SEQ\_ID NO 14

&lt;211&gt; LENGTH: 252

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 14

Met Ser Leu Leu Thr Glu Val Glu Thr Tyr Val Leu Ser Ile Val Pro			
1	5	10	15

Ser Gly Pro Leu Lys Ala Glu Ile Ala Gln Arg Leu Glu Asp Val Phe			
20	25	30	

Ala Gly Lys Asn Thr Asp Leu Glu Ala Leu Met Glu Trp Leu Lys Thr			
35	40	45	

Arg Pro Ile Leu Ser Pro Leu Thr Lys Gly Ile Leu Gly Phe Val Phe			
50	55	60	

Thr Leu Thr Val Pro Ser Glu Arg Gly Leu Gln Arg Arg Arg Phe Val			
65	70	75	80

Gln Asn Ala Leu Asn Gly Asn Gly Asp Pro Asn Asn Met Asp Lys Ala			
85	90	95	

Val Lys Leu Tyr Arg Lys Leu Lys Arg Glu Ile Thr Phe His Gly Ala			
100	105	110	

Lys Glu Val Ala Leu Ser Tyr Ser Ala Gly Ala Leu Ala Ser Cys Met			
115	120	125	

Gly Leu Ile Tyr Asn Arg Met Gly Thr Val Ala Thr Glu Val Ala Phe			
130	135	140	

Gly Leu Val Cys Ala Thr Cys Glu Gln Ile Ala Asp Ser Gln His Arg			
145	150	155	160

Ser His Arg Gln Met Val Thr Thr Asn Pro Leu Ile Arg His Glu			
165	170	175	

Asn Arg Met Val Ile Ala Ser Thr Thr Ala Lys Ala Met Glu Gln Met			
180	185	190	

Ala Gly Ser Ser Glu Gln Ala Ala Glu Ala Met Glu Val Ala Ser Gln			
195	200	205	

Ala Arg Gln Met Val Gln Ala Met Arg Thr Ile Gly Thr His Pro Ser			
210	215	220	

Ser Ser Thr Gly Leu Lys Asp Asp Leu Leu Glu Asn Leu Gln Ala Tyr			
225	230	235	240

Gln Lys Arg Met Gly Val Gln Met Gln Arg Phe Lys			
245	250		

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<210> SEQ ID NO 15
<211> LENGTH: 657
<212> TYPE: DNA
<213> ORGANISM: Influenza A virus

<400> SEQUENCE: 15

atggactcca atactgtgtc aagctttcag gtagactgtt tcctttggca catccgcaaa      60
cgatttgcag acaatggatt gggtgatgcc ccattccttg atcggctccg ccgagatcaa     120
aagtccctaa aaggaaagggg caaacccctt agcctagaca tcgaaaacagc cactttgtt     180
gggaaacaaa ttgttgagtg gatttgaaa gaggaatcca gcgatatact taagatgacc     240
attgcacatcg tgcctacttc ggcgttaccta gctgacatga ccctcgagga aatgtcacga   300
gactggttca tgctaatgcc taggcaaaag ataataggcc ctctttgtt gcgagtggac   360
caggcgtatca tggaaaagaa catcatactg aaagcgaact tcagtgatgc cttaaccga   420
tttagagactt tgatactact aagggtttc actgaggagg gagcaatcgt tggagaatt   480
tcaccattac ottatcttcc aggacatact aatgaggatg taaaaatgc agttgggtc   540
ctcatcgag ggcttgaatg gaatggtaac acggttcgag gctctgaaaa tctacagaga   600
ttcgcttggaa gaaaccataa tgaggatggg agatcttcac tacctccaga acagaaa   657

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<210> SEQ ID NO 16
<211> LENGTH: 219
<212> TYPE: PRT
<213> ORGANISM: Influenza A virus

<400> SEQUENCE: 16

Met Asp Ser Asn Thr Val Ser Ser Phe Gln Val Asp Cys Phe Leu Trp
1           5          10          15

His Ile Arg Lys Arg Phe Ala Asp Asn Gly Leu Gly Asp Ala Pro Phe
20          25          30

Leu Asp Arg Leu Arg Arg Asp Gln Lys Ser Leu Lys Gly Arg Gly Asn
35          40          45

Thr Leu Ser Leu Asp Ile Glu Thr Ala Thr Leu Val Gly Lys Gln Ile
50          55          60

Val Glu Trp Ile Leu Lys Glu Glu Ser Ser Asp Ile Leu Lys Met Thr
65          70          75          80

Ile Ala Ser Val Pro Thr Ser Arg Tyr Leu Ala Asp Met Thr Leu Glu
85          90          95

Glu Met Ser Arg Asp Trp Phe Met Leu Met Pro Arg Gln Lys Ile Ile
100         105         110

Gly Pro Leu Cys Val Arg Val Asp Gln Ala Ile Met Glu Lys Asn Ile
115         120         125

Ile Leu Lys Ala Asn Phe Ser Val Ile Phe Asn Arg Leu Glu Thr Leu
130         135         140

Ile Leu Leu Arg Ala Phe Thr Glu Glu Gly Ala Ile Val Gly Glu Ile
145         150         155         160

Ser Pro Leu Pro Tyr Leu Pro Gly His Thr Asn Glu Asp Val Lys Asn
165         170         175

Ala Val Gly Val Leu Ile Gly Gly Leu Glu Trp Asn Gly Asn Thr Val
180         185         190

Arg Gly Ser Glu Asn Leu Gln Arg Phe Ala Trp Arg Asn His Asn Glu
195         200         205

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Asp Gly Arg Ser Ser Leu Pro Pro Glu Gln Lys
210         215

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<210> SEQ ID NO 17  
<211> LENGTH: 1695  
<212> TYPE: DNA  
<213> ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 17

atgaaagtaa aactaatggt tctgttatgt acatttacag ctacatatgc agacacaata	60
tgtgtaggct accatgccaa caactcaact gacactgttg acacagtact tgagaagaat	120
gtgacagtga cacactctgt caacctactt gaggacagcc acaatggaaa actatgtcta	180
ctaaaaggaa tagctccact acaattgggt agttgcagcg ttgcggatg gatcttagga	240
aacccagagt gcgaattgct gatttccaag gaatcttggt cctacattgt agaaacacca	300
aatcctgaga atggAACATG ttaccCAGGG tatttcACAG actatGAAGA actgaggGAG	360
caatttGAGTT cAGTATCTTC atttaAGAGG ttCGAAATAT tCCCCAAAGA gagCTCATGG	420
CCCAACCACA CCGTAACCGG AGTGTCACTCA TCACTGCTCC ATAACGGGAA AAGCAGCTC	480
TACAGAAATT TGCTATGGCT GACGGTGAAG AACGGTCTGT ACCCAAACCT GAGCAAGTCC	540
TATACAAACA AAAAGGAGAA AGAAGTCCTT GTACTATGGG GTGTTCACTCA CCCATCTAAC	600
ATAGGGGACC AAAGGGCCCT CTATCATACA GAAAATGCTT ATGTCTCTGT AGTGTCTTC	660
CATTATAGCA GAAGATTCA CCCCAGAAATA GCCAAAAGAC CCAAGGTGAG AAATCAGGAA	720
GGAAGAAATCA ACTACTACTG GACCCCTGCTA GAACCCGGGG ATACAATAAT ATTGAGGCA	780
AATGGAAATC TAATAGCACC AAGGTATGCC TTCGAACTGA GTAAGGGTT TGGATCAGGA	840
ATCATCACAT CAAATGCACC AATGGGTGAA TGTAAATGCAA AGTGTCAAAC ACCTCAGGGA	900
GCTATAAACCA GCAGTCTTCC TTTCCAGAAAT GTACACCCAG TAACAATAGG AGAGTGCCC	960
AAGTATGTCA AAAGTCAAA ATTAAGGTGTT GTTACAGGAC TAAGGAACAC CCCATCCATT	1020
CAATCCAGAG GTTGTGTTGG AGCCATTGCC GGTTTCATTG AAGGAGGGTG GACTGGAATG	1080
GTAGATGGTT GGTATGGTTA TCACCATCAG AATGAGCAAG GATCTGGGTA TGCTGCAGAC	1140
CAACAAAGCA CACAAATGC CATTATGGG ATTACAAACA AGGTGAATTG TGTGATTGAA	1200
AAAATGAACA CTCATTACAC AGCTGTGGGC AAAGAATTCA ACAAACTGGA AAGAAGAATG	1260
AAAAACTTAA ATAAAAAGGT TGTATGATGGG TTTCTAGACA TTGGACATA TAATGCAGAA	1320
TTGTTAGTTC TACTGGAAAAA TGAAAGGACT TTGGATTCC ATGACTCCAA CGTGAAGAAT	1380
CTGTATGAGA AAGTAAAAAG CCAATTAAAA AATAATGCCA AAGAAATAGG AAACGGGTGT	1440
TTTGAATTCT ATCATAAGTG TAACGATGAA TGCATGGAGA GTGTGAAAAA TTGGAACTTAT	1500
GACTATCCAA AATATTCCGA AGAATCAAAG TAAACAGGG AGAAAATTGA TTGGAGTGA	1560
TTGGAATCAA TGGGAGTCTA TAATATCCTG GCGATCTACT CAACAGTCGC CAGTCCCTA	1620
GTTCCTTCTAG TCTCCCTGGG GGCAATCAGC TTCTGGATGT GTTCCAATGG GTCTTACAG	1680
TGTAGAATAT GCACTC	1695

<210> SEQ ID NO 18  
<211> LENGTH: 565  
<212> TYPE: PRT  
<213> ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 18

Met Lys Val Lys Leu Met Val Leu Leu Cys Thr Phe Thr Ala Thr Tyr	
1 5 10 15	
Ala Asp Thr Ile Cys Val Gly Tyr His Ala Asn Asn Ser Thr Asp Thr	
20 25 30	

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Val Asp Thr Val Leu Glu Lys Asn Val Thr Val Thr His Ser Val Asn  
   35                  40                  45  
 Leu Leu Glu Asp Ser His Asn Gly Lys Leu Cys Leu Leu Lys Gly Ile  
   50                  55                  60  
 Ala Pro Leu Gln Leu Gly Ser Cys Ser Val Ala Gly Trp Ile Leu Gly  
   65                  70                  75                  80  
 Asn Pro Glu Cys Glu Leu Leu Ile Ser Lys Glu Ser Trp Ser Tyr Ile  
   85                  90                  95  
 Val Glu Thr Pro Asn Pro Glu Asn Gly Thr Cys Tyr Pro Gly Tyr Phe  
   100                105                110  
 Thr Asp Tyr Glu Glu Leu Arg Glu Gln Leu Ser Ser Val Ser Ser Phe  
   115                120                125  
 Lys Arg Phe Glu Ile Phe Pro Lys Glu Ser Ser Trp Pro Asn His Thr  
   130                135                140  
 Val Thr Gly Val Ser Ser Ser Cys Ser His Asn Gly Lys Ser Ser Phe  
   145                150                155                160  
 Tyr Arg Asn Leu Leu Trp Leu Thr Val Lys Asn Gly Leu Tyr Pro Asn  
   165                170                175  
 Leu Ser Lys Ser Tyr Thr Asn Lys Lys Glu Lys Glu Val Leu Val Leu  
   180                185                190  
 Trp Gly Val His His Pro Ser Asn Ile Gly Asp Gln Arg Ala Leu Tyr  
   195                200                205  
 His Thr Glu Asn Ala Tyr Val Ser Val Val Ser Ser His Tyr Ser Arg  
   210                215                220  
 Arg Phe Thr Pro Glu Ile Ala Lys Arg Pro Lys Val Arg Asn Gln Glu  
   225                230                235                240  
 Gly Arg Ile Asn Tyr Tyr Trp Thr Leu Leu Glu Pro Gly Asp Thr Ile  
   245                250                255  
 Ile Phe Glu Ala Asn Gly Asn Leu Ile Ala Pro Arg Tyr Ala Phe Glu  
   260                265                270  
 Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Pro Met  
   275                280                285  
 Gly Glu Cys Asn Ala Lys Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
   290                295                300  
 Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
   305                310                315                320  
 Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
   325                330                335  
 Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
   340                345                350  
 Ile Glu Gly Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
   355                360                365  
 His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Gln Gln Ser Thr  
   370                375                380  
 Gln Asn Ala Ile Asn Gly Ile Thr Asn Lys Val Asn Ser Val Ile Glu  
   385                390                395                400  
 Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Asn Lys Leu  
   405                410                415  
 Glu Arg Arg Met Glu Asn Leu Asn Lys Lys Val Asp Asp Gly Phe Leu  
   420                425                430  
 Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn Glu  
   435                440                445

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Arg	Thr	Leu	Asp	Phe	His	Asp	Ser	Asn	Val	Lys	Asn	Leu	Tyr	Glu	Lys
450						455				460					

Val	Lys	Ser	Gln	Leu	Lys	Asn	Asn	Ala	Lys	Glu	Ile	Gly	Asn	Gly	Cys
465					470				475				480		

Phe	Glu	Phe	Tyr	His	Lys	Cys	Asn	Asp	Glu	Cys	Met	Glu	Ser	Val	Lys
485						490				495					

Asn	Gly	Thr	Tyr	Asp	Tyr	Pro	Lys	Tyr	Ser	Glu	Glu	Ser	Lys	Leu	Asn
500						505				510					

Arg	Glu	Ile	Asp	Gly	Val	Lys	Leu	Glu	Ser	Met	Gly	Val	Tyr	Asn
515					520				525					

Ile	Leu	Ala	Ile	Tyr	Ser	Thr	Val	Ala	Ser	Ser	Leu	Val	Leu	Leu	Val
530					535				540						

Ser	Leu	Gly	Ala	Ile	Ser	Phe	Trp	Met	Cys	Ser	Asn	Gly	Ser	Leu	Gln
545					550				555				560		

Cys	Arg	Ile	Cys	Ile											
				565											

&lt;210&gt; SEQ\_ID NO 19

&lt;211&gt; LENGTH: 1407

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Influenza A virus

&lt;400&gt; SEQUENCE: 19

atgaatccaa	atcaaaggat	aataacgatt	ggctctgttt	ctcttactat	tgccacaatg	60
tgcttcctta	tgcaaattgc	cattctggta	actaatgtaa	cattgcactt	caatcaatat	120
gaatgcact	ccccccaaa	caaccaagta	atactgtgt	aaccaacaat	aatagaaaga	180
aacataacag	agatagtgt	tctgaccaac	accaccatag	agaaggaaat	atgccccaaa	240
ctagcagaat	acagaaattg	gtcaaagccg	caatgtaaaa	ttacagggtt	tgcaccttt	300
tccaaggaca	attcgattag	gcttctgtct	ggtggggaca	tttgggtgac	gagagaacct	360
tatgtgtcat	gcgatcctga	taagtgttat	cagtttgc	ttggacaagg	aacaacattta	420
aacaacaggc	attcaaatga	cacagtacat	gataggaccc	cttacgaaac	cctattgtat	480
aatgagttgg	gtattccatt	ccatTTGGGG	accaaacaag	tgtgcata	atggtccagc	540
tcaagttgtc	atgatggaaa	agcatggctt	cacgtttgt	ttactggca	tgattraaaat	600
geaactgcta	gcttcattta	caatgggggg	ctttagata	gtattgggtc	atggtccaaa	660
aaaatactca	ggacacagga	gtcggaatgt	gtttgcata	atggaaactg	tacagtagta	720
atgactgatg	ggagtgc	tttc	aggaatagct	gacactaaaa	tattattcat	780
aaaatcggtc	atattagccy	attgttagga	agtgc	tgc	atgttagagga	840
tatccccgt	atcctgggt	cagatgc	tgt	agaca	actggaaagg	900
cccgtcgtag	atataaatgt	aaaggattat	agcattgttt	ccagttatgt	gtgc	960
cttggggag	atacacc	aaaagacgac	agatcc	cgat	tctgaaatc	1020
aacaatgagg	aaggggggca	tggagtgaaa	ggctgggc	ttgatgatgg	aaatgatgt	1080
tggatggggaa	gaacaatcaa	cgagacgtt	cgctcaggtt	atgaaac	caaagtcat	1140
gaaggctgg	ccaaacctaa	ttccaaattt	cagataaata	ggcaagtc	atgtgaaaga	1200
ggtgataggt	ccggttattt	tggcatttt	tctgttgaag	gcaaaagctg	tatcaatcg	1260
tgctttatg	tggagttgat	aagaggaagg	aaacaggaaa	ctgc	atgtgaaaga	1320
aacagtattt	ttgtgtttt	tggcacctca	ggtacat	gaa	acaggctc	1380
ggggcgaaca	tcaatctcat	gcctgtta				1407

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<210> SEQ\_ID NO 20  
 <211> LENGTH: 469  
 <212> TYPE: PRT  
 <213> ORGANISM: Influenza A virus  
 <220> FEATURE:  
 <221> NAME/KEY: misc\_feature  
 <222> LOCATION: (199)...(199)  
 <223> OTHER INFORMATION: Xaa can be any naturally occurring amino acid  
 <220> FEATURE:  
 <221> NAME/KEY: misc\_feature  
 <222> LOCATION: (267)...(267)  
 <223> OTHER INFORMATION: Xaa can be any naturally occurring amino acid

&lt;400&gt; SEQUENCE: 20

Met	Asn	Pro	Asn	Gln	Lys	Ile	Ile	Thr	Ile	Gly	Ser	Val	Ser	Leu	Thr
1						5		10					15		

Ile	Ala	Thr	Met	Cys	Phe	Leu	Met	Gln	Ile	Ala	Ile	Leu	Val	Thr	Asn
						20		25				30			

Val	Thr	Leu	His	Phe	Asn	Gln	Tyr	Glu	Cys	Asn	Tyr	Pro	Pro	Asn	Asn
						35		40			45				

Gln	Val	Ile	Leu	Cys	Glu	Pro	Thr	Ile	Ile	Glu	Arg	Asn	Ile	Thr	Glu
						50		55		60					

Ile	Val	Tyr	Leu	Thr	Asn	Thr	Thr	Ile	Glu	Lys	Glu	Ile	Cys	Pro	Lys
						65		70		75		80			

Leu	Ala	Glu	Tyr	Arg	Asn	Trp	Ser	Lys	Pro	Gln	Cys	Lys	Ile	Thr	Gly
						85		90		95					

Phe	Ala	Pro	Phe	Ser	Lys	Asp	Asn	Ser	Ile	Arg	Leu	Ser	Ala	Gly	Gly
						100		105		110					

Asp	Ile	Trp	Val	Thr	Arg	Glu	Pro	Tyr	Val	Ser	Cys	Asp	Pro	Asp	Lys
						115		120		125					

Cys	Tyr	Gln	Phe	Ala	Leu	Gly	Gln	Gly	Thr	Thr	Leu	Asn	Asn	Arg	His
						130		135		140					

Ser	Asn	Asp	Thr	Val	His	Asp	Arg	Thr	Pro	Tyr	Arg	Thr	Leu	Leu	Met
						145		150		155		160			

Asn	Glu	Leu	Gly	Ile	Pro	Phe	His	Leu	Gly	Thr	Lys	Gln	Val	Cys	Ile
						165		170		175					

Ala	Trp	Ser	Ser	Ser	Cys	His	Asp	Gly	Lys	Ala	Trp	Leu	His	Val	
						180		185		190					

Cys	Ile	Thr	Gly	His	Asp	Xaa	Asn	Ala	Thr	Ala	Ser	Phe	Ile	Tyr	Asn
						195		200		205					

Gly	Arg	Leu	Val	Asp	Ser	Ile	Gly	Ser	Trp	Ser	Lys	Lys	Ile	Leu	Arg
						210		215		220					

Thr	Gln	Glu	Ser	Glu	Cys	Val	Cys	Ile	Asn	Gly	Thr	Cys	Thr	Val	Val
						225		230		235		240			

Met	Thr	Asp	Gly	Ser	Ala	Ser	Gly	Ile	Ala	Asp	Thr	Lys	Ile	Leu	Phe
						245		250		255					

Ile	Glu	Gly	Lys	Ile	Val	His	Ile	Ser	Xaa	Leu	Leu	Gly	Ser	Ala	
						260		265		270					

Gln	His	Val	Glu	Glu	Cys	Ser	Cys	Tyr	Pro	Arg	Tyr	Pro	Gly	Val	Arg
						275		280		285					

Cys	Ile	Cys	Arg	Asp	Asn	Trp	Lys	Gly	Ser	Asn	Arg	Pro	Val	Val	Asp
						290		295		300					

Ile	Asn	Val	Lys	Asp	Tyr	Ser	Ile	Val	Ser	Ser	Tyr	Val	Cys	Ser	Gly
						305		310		315		320			

Leu	Val	Gly	Asp	Thr	Pro	Arg	Lys	Asp	Asp	Arg	Ser	Ser	Ser	Asp	
						325		330		335					

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Cys Leu Asn Pro Asn Asn Glu Glu Gly Gly His Gly Val Lys Gly Trp  
                  340                   345                   350

Ala Phe Asp Asp Gly Asn Asp Val Trp Met Gly Arg Thr Ile Asn Glu  
355 360 365

Thr Leu Arg Ser Gly Tyr Glu Thr Phe Lys Val Ile Glu Gly Trp Ser  
 370                    375                    380

Lys Pro Asn Ser Lys Leu Gln Ile Asn Arg Gln Val Ile Val Glu Arg  
385 390 395 400

Gly Asp Arg Ser Gly Tyr Ser Gly Ile Phe Ser Val Glu Gly Lys Ser  
405 410 415

Cys Ile Asn Arg Cys Phe Tyr Val Glu Leu Ile Arg Gly Arg Lys Gln  
 420 425 430

Glu Thr Ala Val Trp Trp Thr Ser Asn Ser Ile Val Val Phe Cys Gly  
435 440 445

Thr Ser Gly Thr Tyr Gly Thr Gly Ser Trp Pro Asp Gly Ala Asn Ile  
450 455 460

Asn Leu Met Pro Val  
465

<210> SEQ ID NO 21

<21

<210> SEQ ID NO 21  
<211> LENGTH: 1695  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: HA gene of Influenza 10-0036-2 (n+5, relative  
to parent)

<400> SEQUENCE: 21

atgaaaatgtt aactaatggc tctgttatgt acatttacag ctacatatgc agacacaata 60  
tgtgttaggct accatgccaa caactcaact gacactgtt acacagtact tgagaagaat 120  
gtgacagtga cacactctgt caacctactt gaggacagcc acaatggaaa actatgtcta 180  
ctaaaaggaa tagctccact acaattgggt aattgcagcg ttgcggatg gatcttagga 240  
aaccaggat gcgaattgtt gattccaat gaatcttggtt cctacattgtt agaaacacca 300  
aatcctgaga atgaaacatg ttacccaggg tatttcacag actatgaaga actgaggag 360  
caatttgaggat cagtatcttc atttaagagg ttgcataat tccccaaaga gagctcatgg 420  
cccaaccaca ccgtaacccgg agtgtcatca tcatgctccc ataacggaa aagcagcttc 480  
tacagaaatt tgctatggct gacgggttgaag aacgggttactt acccaaacctt gagcaagtcc 540  
tatacaaaca aaaaggagaa agaagtcctt gtactatggg gtgttcatca cccatctaacc 600  
ataggggacc aaagggccct ctatcataca gaaaatgtttt atgtctctgtt agtgtttca 660  
cattatagca gaagattcac cccagaaata gccaaaagac ccaagggtttagaa aatcaggaa 720  
ggaagaatca actactactg gaccctgttta gaacccgggg atacaataat atttgaggca 780  
aatggaaatc taatagcacc aaggtatgtt ttcgaacttga gtaagggtttt tggatcaggaa 840  
atcatcacat ccaaataatcaac aatgggttgaat gttatgttca cgtgtttttt acctcaggaa 900  
gttataaaaca gcaatgtttcc ttccagaat gtacacccag taacaatagg agagtgcacca 960  
aagtatgtca aaagtgcacaa attaaggatg gttacaggac taaggaacac cccatccattt 1020  
caatccagag gtttgggg agccattgttcc ggttttcatgtt aaggagggtt gacttggaaatg 1080  
gttagatgggtt ggtatgggtt tcaccatgtt aatggatgtt gatctgggtt tgctgtttttt 1140  
caacaaagca cacaaaaatgc cattaatggg attacaaaca aggtgttacccatgtt tggatgtt 1200

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aaaatgaaca ctcaattcac agctgtggc aaagaattca acaaactgga aagaagaatg 1260
aaaaacttaa ataaaaaggt tgatgtggg tttctagaca tttggacata taatgcagaa 1320
ttgttagttc tactggaaaa tcaaaggact ttggattcc atgactccaa cgtgaagaat 1380
ctgttatgaga aagtaaaaag ccaattaaaa aataatgcc aagaaatagg aaacgggtgt 1440
tttgaattct atcataagtg taacgatgaa tgcattggaga gtgtgaaaaa tggacttat 1500
gactatccaa aatattccga agaatcaaag ttaaacaggg agaaaattga tggagtgaaa 1560
ttggaatcaa tgggagtcta taatatcctg gcatctact caacagtgc cagttcccta 1620
gttcttttag tctccctggg ggcaatcagc ttctggatgt gttccaatgg gtctttacag 1680
tgtagaatat gcatac 1695

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&lt;210&gt; SEQ\_ID NO 22

&lt;211&gt; LENGTH: 565

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Artificial Sequence

&lt;220&gt; FEATURE:

<223> OTHER INFORMATION: HA predicted AA of Influenza 10-0036-2 (n+5,  
relative to parent)

&lt;400&gt; SEQUENCE: 22

Met	Lys	Val	Lys	Leu	Met	Val	Leu	Leu	Cys	Thr	Phe	Thr	Ala	Thr	Tyr
1									10						15

Ala	Asp	Thr	Ile	Cys	Val	Gly	Tyr	His	Ala	Asn	Asn	Ser	Thr	Asp	Thr
			20						25				30		

Val	Asp	Thr	Val	Leu	Glu	Lys	Asn	Val	Thr	Val	Thr	His	Ser	Val	Asn
					35				40			45			

Leu	Leu	Glu	Asp	Ser	His	Asn	Gly	Lys	Leu	Cys	Leu	Leu	Lys	Gly	Ile
					50				55			60			

Ala	Pro	Leu	Gln	Leu	Gly	Asn	Cys	Ser	Val	Ala	Gly	Trp	Ile	Leu	Gly
					65				70			75			80

Asn	Pro	Glu	Cys	Glu	Leu	Leu	Ile	Ser	Asn	Glu	Ser	Trp	Ser	Tyr	Ile
					85				90			95			

Val	Glu	Thr	Pro	Asn	Pro	Glu	Asn	Gly	Thr	Cys	Tyr	Pro	Gly	Tyr	Phe
					100				105			110			

Thr	Asp	Tyr	Glu	Glu	Leu	Arg	Glu	Gln	Leu	Ser	Ser	Val	Ser	Ser	Phe
					115				120			125			

Lys	Arg	Phe	Glu	Ile	Phe	Pro	Lys	Glu	Ser	Ser	Trp	Pro	Asn	His	Thr
					130				135			140			

Val	Thr	Gly	Val	Ser	Ser	Ser	Cys	Ser	His	Asn	Gly	Lys	Ser	Ser	Phe
					145				150			155			160

Tyr	Arg	Asn	Leu	Leu	Trp	Leu	Thr	Val	Lys	Asn	Gly	Thr	Tyr	Pro	Asn
					165				170			175			

Leu	Ser	Lys	Ser	Tyr	Thr	Asn	Lys	Lys	Glu	Lys	Val	Leu	Val	Leu	
					180				185			190			

Trp	Gly	Val	His	His	Pro	Ser	Asn	Ile	Gly	Asp	Gln	Arg	Ala	Leu	Tyr
					195				200			205			

His	Thr	Glu	Asn	Ala	Tyr	Val	Ser	Val	Val	Ser	Ser	His	Tyr	Ser	Arg
					210				215			220			

Arg	Phe	Thr	Pro	Glu	Ile	Ala	Lys	Arg	Pro	Lys	Val	Arg	Asn	Gln	Glu
					225				230			235			240

Gly	Arg	Ile	Asn	Tyr	Tyr	Trp	Thr	Leu	Leu	Glu	Pro	Gly	Asp	Thr	Ile
					245				250			255			

Ile	Phe	Glu	Ala	Asn	Gly	Asn	Leu	Ile	Ala	Pro	Arg	Tyr	Ala	Phe	Glu
					260				265			270			

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Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Thr Met  
275 280 285

Gly Glu Cys Asn Ala Thr Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
290 295 300

Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
305 310 315 320

Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
325 330 335

Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
340 345 350

Ile Glu Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
355 360 365

His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Gln Gln Ser Thr  
370 375 380

Gln Asn Ala Ile Asn Gly Ile Thr Asn Lys Val Asn Ser Val Ile Glu  
385 390 395 400

Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Asn Lys Leu  
405 410 415

Glu Arg Arg Met Glu Asn Leu Asn Lys Lys Val Asp Asp Gly Phe Leu  
420 425 430

Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn Glu  
435 440 445

Arg Thr Leu Asp Phe His Asp Ser Asn Val Lys Asn Leu Tyr Glu Lys  
450 455 460

Val Lys Ser Gln Leu Lys Asn Asn Ala Lys Glu Ile Gly Asn Gly Cys  
465 470 475 480

Phe Glu Phe Tyr His Lys Cys Asn Asp Glu Cys Met Glu Ser Val Lys  
485 490 495

Asn Gly Thr Tyr Asp Tyr Pro Lys Tyr Ser Glu Glu Ser Lys Leu Asn  
500 505 510

Arg Glu Lys Ile Asp Gly Val Lys Leu Glu Ser Met Gly Val Tyr Asn  
515 520 525

Ile Leu Ala Ile Tyr Ser Thr Val Ala Ser Ser Leu Val Leu Val  
530 535 540

Ser Leu Gly Ala Ile Ser Phe Trp Met Cys Ser Asn Gly Ser Leu Gln  
545 550 555 560

Cys Arg Ile Cys Ile  
565

<210> SEQ ID NO 23

<211> LENGTH: 1695

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: HA gene of Influenza 10-0036-2 (n+4, relative to parent)

<400> SEQUENCE: 23

atgaaaagtaa aactaatgggt tctgttatgt acatttacag ctacatatgc agacacaata 60

tgtgtaggct accatgccaa caactcaact gacactgttg acacagtact tgagaagaat 120

gtgacagtga cacactctgt caacctactt gaggacagcc acaatggaaa actatgtcta 180

ctaaaaggaa tagctccact acaattgggt aattgcagcg ttgcccggatg gatcttagga 240

aacccagagt gcgaattgct gatttccaat gaatcttgggt cctacattgt agaaacacca 300

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aatcctgaga atggaacatg ttacccaggg tatttcacag actatgaaga actgaggagg	360
caattgagtt cagtatctc attaagagg ttcgaaatat tcccaaaga gagctcatgg	420
cccaaccaca ccgtAACCGG agtgcata tcatgctccc ataacggaa aagcagctc	480
tacagaaatt tgctatggct gacggtaag aacggtaatg acccaaacct gagcaagtcc	540
tatacaaaca aaaaggagaa agaagtccct gtactatggg gtgttcatca cccatctaacc	600
atagggacc aaagggccct ctatcataca gaaaatgctt atgtctctgt agtgtctca	660
cattatagca gaagattcac cccagaaata gccaaaagac ccaaggtagg aaatcaggaa	720
ggaagaatca actactactg gaccctgcta gaacccgggg atacaataat atttgaggca	780
aatggaaatc taatagcacc aaggtagtgc ttgcactga gtaagggttt tggatcagga	840
atcatcacat caaatgcaac aatgggtaa tgtaatgcaa agtgtcaaac acctcaggaa	900
gctataaaca gcagtcttcc tttccagaat gtacacccag taacaatagg agagtgcacca	960
aagtatgtca aagtgcaaa attaaggatg gttacaggac taaggaacac cccatccatt	1020
caatccagag gtttggggg agccattgcc gggttcatgg aaggagggtg gactggaaatg	1080
gtagatgggtt ggtatggta tcaccatcg aatgagcaag gatctgggtt tgctgcagac	1140
caacaaagca cacaaaatgc cattaatggg attacaaaca aggtgaatc tgtgattgaa	1200
aaaaatgaaca ctcaattcac agctgtgggc aaagaattca acaaactgga aagaagaatg	1260
aaaaacttaa ataaaaaggt tgatgatggg tttctagaca ttggacata taatgcagaa	1320
ttgttagttc tactggaaaa taaaaggact ttggatttcc atgactccaa cgtgaagaat	1380
ctgtatgaga aagtaaaaag ccaattaaaa aataatgcc aagaaatagg aaacgggtgt	1440
tttgaattct atcataagtg taacgatgaa tgcattggaa gtgtgaaaaa ttggacttat	1500
gactatccaa aatattccga agaatcaaag taaaacaggg agaaaattga ttggagtggaa	1560
tttggatcaa tggagttca taatatcctg gcgttactt caacagtgcg cagttcccta	1620
gttcttttag tctccctggg ggcattcgc ttctggatgt gttccaatgg gtctttacag	1680
tgttagatat gcattc	1695

&lt;210&gt; SEQ ID NO 24

&lt;211&gt; LENGTH: 565

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Artificial Sequence

&lt;220&gt; FEATURE:

&lt;223&gt; OTHER INFORMATION: HA predicted AA of Influenza 10-0036-2 (n+4, relative to parent)

&lt;400&gt; SEQUENCE: 24

Met Lys Val Lys Leu Met Val Leu Leu Cys Thr Phe Thr Ala Thr Tyr			
1	5	10	15

Ala Asp Thr Ile Cys Val Gly Tyr His Ala Asn Asn Ser Thr Asp Thr			
20	25	30	

Val Asp Thr Val Leu Glu Lys Asn Val Thr Val Thr His Ser Val Asn			
35	40	45	

Leu Leu Glu Asp Ser His Asn Gly Lys Leu Cys Leu Leu Lys Gly Ile			
50	55	60	

Ala Pro Leu Gln Leu Gly Asn Cys Ser Val Ala Gly Trp Ile Leu Gly			
65	70	75	80

Asn Pro Glu Cys Glu Leu Leu Ile Ser Asn Glu Ser Trp Ser Tyr Ile			
85	90	95	

Val Glu Thr Pro Asn Pro Glu Asn Gly Thr Cys Tyr Pro Gly Tyr Phe			
100	105	110	

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Thr Asp Tyr Glu Glu Leu Arg Glu Gln Leu Ser Ser Val Ser Ser Phe  
 115 120 125  
 Lys Arg Phe Glu Ile Phe Pro Lys Glu Ser Ser Trp Pro Asn His Thr  
 130 135 140  
 Val Thr Gly Val Ser Ser Ser Cys Ser His Asn Gly Lys Ser Ser Phe  
 145 150 155 160  
 Tyr Arg Asn Leu Leu Trp Leu Thr Val Lys Asn Gly Thr Tyr Pro Asn  
 165 170 175  
 Leu Ser Lys Ser Tyr Thr Asn Lys Lys Glu Lys Glu Val Leu Val Leu  
 180 185 190  
 Trp Gly Val His His Pro Ser Asn Ile Gly Asp Gln Arg Ala Leu Tyr  
 195 200 205  
 His Thr Glu Asn Ala Tyr Val Ser Val Val Ser Ser His Tyr Ser Arg  
 210 215 220  
 Arg Phe Thr Pro Glu Ile Ala Lys Arg Pro Lys Val Arg Asn Gln Glu  
 225 230 235 240  
 Gly Arg Ile Asn Tyr Tyr Trp Thr Leu Leu Glu Pro Gly Asp Thr Ile  
 245 250 255  
 Ile Phe Glu Ala Asn Gly Asn Leu Ile Ala Pro Arg Tyr Ala Phe Glu  
 260 265 270  
 Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Thr Met  
 275 280 285  
 Gly Glu Cys Asn Ala Lys Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
 290 295 300  
 Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
 305 310 315 320  
 Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
 325 330 335  
 Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
 340 345 350  
 Ile Glu Gly Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
 355 360 365  
 His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Gln Gln Ser Thr  
 370 375 380  
 Gln Asn Ala Ile Asn Gly Ile Thr Asn Lys Val Asn Ser Val Ile Glu  
 385 390 395 400  
 Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Asn Lys Leu  
 405 410 415  
 Glu Arg Arg Met Glu Asn Leu Asn Lys Lys Val Asp Asp Gly Phe Leu  
 420 425 430  
 Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn Glu  
 435 440 445  
 Arg Thr Leu Asp Phe His Asp Ser Asn Val Lys Asn Leu Tyr Glu Lys  
 450 455 460  
 Val Lys Ser Gln Leu Lys Asn Asn Ala Lys Glu Ile Gly Asn Gly Cys  
 465 470 475 480  
 Phe Glu Phe Tyr His Lys Cys Asn Asp Glu Cys Met Glu Ser Val Lys  
 485 490 495  
 Asn Gly Thr Tyr Asp Tyr Pro Lys Tyr Ser Glu Glu Ser Lys Leu Asn  
 500 505 510  
 Arg Glu Lys Ile Asp Gly Val Lys Leu Glu Ser Met Gly Val Tyr Asn  
 515 520 525

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Ile Leu Ala Ile Tyr Ser Thr Val Ala Ser Ser Leu Val Leu Leu Val  
530 535 540

Ser Leu Gly Ala Ile Ser Phe Trp Met Cys Ser Asn Gly Ser Leu Gln  
545 550 555 560

Cys Arg Ile Cys Ile  
565

<210> SEQ\_ID NO 25

<211> LENGTH: 1695

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: HA gene of Influenza 10-0036-2 (n+3, relative to parent)

<400> SEQUENCE: 25

atgaaaagtaa aactaatggc	tctgttatgt acatttacag	ctacatatgc agacacaata	60
tgtgtaggct accatgccaa caactcaact	gacactgttg acacagtact	tgagaagaat	120
gtgacagtga cacaactctgt	caacctactt gaggacagcc	acaatggaaa actatgtcta	180
ctaaaaggaa tagctccact	acaattgggt aattgcagcg	ttgccccatg gatcttagga	240
aacccagagt gcgaattgct	gatttccat gaatcttggt	cctacattgt agaaaacacca	300
aatcctgaga atgaaacatg	ttacccaggg tatttcacag	actatgaaga actgaggag	360
caatttggat tt ctttgcattt	tttgcattttt tcccaaaaga	gagctcatgg	420
cccaaccaca ccgttaaccgg	agtgtcatca tcatgctccc	ataacggaa aagcagcttc	480
tacagaaatt tgctatggct	gacgggtgaag aacgggtacgt	acccaaacct gagcaagtcc	540
tatacaaaca aaaaggagaa agaagtccctt	gtactatggg gtgttcatca	cccatctaac	600
ataggggacc aaaggccct ctatcataca	gaaaatgctt atgtctctgt	agtgtcttca	660
cattatagca gaagattcac cccagaaata	gccaaggac ccaagggtgag	aaatcaggaa	720
ggaagaatca actactactg gaccctgcta	gaaccggggg atacaataat	atttgaggca	780
aatggaaatc taatagcacc aaggtatgcc	ttcgaactga gtaagggtt	tggatcagga	840
atcatcacat caaatgcacc aatgggtgaa	tgtatgccaa agtgtcaaac	acctcaggga	900
gtataaaaca gcagtcttcc tttccagaat	gtacacccag taacaatagg	agagtgcaca	960
aagtatgtca aaagtgc当地 attaaggatg	gttacaggac taaggaacac	cccatccatt	1020
caatccagag gtttggggg agccattgcc	ggtttcattt aaggagggtg	gactggaaatg	1080
gttagatgggtt ggtatgggta tcaccatcg	aatggcaag gatctgggtt	tgctgcagac	1140
caacaaagca cacaaaatgc cattaatggg	attacaaaca aggtgaattc	tgtgattgaa	1200
aaaatgaaca ctcaattcac agctgtgggc	aaagaattca acaaactgga	aagaagaatg	1260
aaaaacttaa ataaaaaggc tgatgtggg	tttcttagaca tttggacata	taatgcagaa	1320
ttgttagttc tactggaaaa tggaaaggact	ttggattcc atgactccaa	cgtgaagaat	1380
ctgtatgaga aagtaaaaag ccaattaaa	aataatgcc aagaaatagg	aaacgggtgt	1440
tttgaattct atcataagtg taacgatgaa	tgcattggaga gtgtgaaaaa	tggacttat	1500
gactatccaa aatattccga agaatcaaag	ttaaacaggg agaaaattga	tggagtgaaa	1560
tttggaaatcaa tgggaggctta taatatccctg	gcatctact caacagtgcgc	cagttcccta	1620
gttcttttag tctccctggg ggcaatcgc	ttctggatgt gttccatgg	gttcttacag	1680
tgttagaaatat gcatac			1695

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<210> SEQ ID NO 26  
 <211> LENGTH: 565  
 <212> TYPE: PRT  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: HA predicted AA of Influenza 10-0036-2 (n+3,  
     relative to parent)  
  
 <400> SEQUENCE: 26  
  
 Met Lys Val Lys Leu Met Val Leu Leu Cys Thr Phe Thr Ala Thr Tyr  
   1               5                   10                   15  
  
 Ala Asp Thr Ile Cys Val Gly Tyr His Ala Asn Asn Ser Thr Asp Thr  
   20              25                   30  
  
 Val Asp Thr Val Leu Glu Lys Asn Val Thr Val Thr His Ser Val Asn  
   35              40                   45  
  
 Leu Leu Glu Asp Ser His Asn Gly Lys Leu Cys Leu Leu Lys Gly Ile  
   50              55                   60  
  
 Ala Pro Leu Gln Leu Gly Asn Cys Ser Val Ala Gly Trp Ile Leu Gly  
   65              70                   75                   80  
  
 Asn Pro Glu Cys Glu Leu Leu Ile Ser Asn Glu Ser Trp Ser Tyr Ile  
   85              90                   95  
  
 Val Glu Thr Pro Asn Pro Glu Asn Gly Thr Cys Tyr Pro Gly Tyr Phe  
   100             105                   110  
  
 Thr Asp Tyr Glu Glu Leu Arg Glu Gln Leu Ser Ser Val Ser Ser Phe  
   115             120                   125  
  
 Lys Arg Phe Glu Ile Phe Pro Lys Glu Ser Ser Trp Pro Asn His Thr  
   130             135                   140  
  
 Val Thr Gly Val Ser Ser Cys Ser His Asn Gly Lys Ser Ser Phe  
   145             150                   155                   160  
  
 Tyr Arg Asn Leu Leu Trp Leu Thr Val Lys Asn Gly Thr Tyr Pro Asn  
   165             170                   175  
  
 Leu Ser Lys Ser Tyr Thr Asn Lys Lys Glu Lys Glu Val Leu Val Leu  
   180             185                   190  
  
 Trp Gly Val His His Pro Ser Asn Ile Gly Asp Gln Arg Ala Leu Tyr  
   195             200                   205  
  
 His Thr Glu Asn Ala Tyr Val Ser Val Val Ser Ser His Tyr Ser Arg  
   210             215                   220  
  
 Arg Phe Thr Pro Glu Ile Ala Lys Arg Pro Lys Val Arg Asn Gln Glu  
   225             230                   235                   240  
  
 Gly Arg Ile Asn Tyr Tyr Trp Thr Leu Leu Glu Pro Gly Asp Thr Ile  
   245             250                   255  
  
 Ile Phe Glu Ala Asn Gly Asn Leu Ile Ala Pro Arg Tyr Ala Phe Glu  
   260             265                   270  
  
 Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Pro Met  
   275             280                   285  
  
 Gly Glu Cys Asn Ala Lys Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
   290             295                   300  
  
 Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
   305             310                   315                   320  
  
 Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
   325             330                   335  
  
 Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
   340             345                   350  
  
 Ile Glu Gly Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
   355             360                   365

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His	Gln	Asn	Glu	Gln	Gly	Ser	Gly	Tyr	Ala	Ala	Asp	Gln	Gln	Ser	Thr
370				375					380						
Gln	Asn	Ala	Ile	Asn	Gly	Ile	Thr	Asn	Lys	Val	Asn	Ser	Val	Ile	Glu
385				390				395							400
Lys	Met	Asn	Thr	Gln	Phe	Thr	Ala	Val	Gly	Lys	Glu	Phe	Asn	Lys	Leu
	405					410				415					
Glu	Arg	Arg	Met	Glu	Asn	Leu	Asn	Lys	Lys	Val	Asp	Asp	Gly	Phe	Leu
	420				425					430					
Asp	Ile	Trp	Thr	Tyr	Asn	Ala	Glu	Leu	Leu	Val	Leu	Glu	Asn	Glu	
	435				440					445					
Arg	Thr	Leu	Asp	Phe	His	Asp	Ser	Asn	Val	Lys	Asn	Leu	Tyr	Glu	Lys
	450				455			460							
Val	Lys	Ser	Gln	Leu	Lys	Asn	Asn	Ala	Lys	Glu	Ile	Gly	Asn	Gly	Cys
465					470				475			480			
Phe	Glu	Phe	Tyr	His	Lys	Cys	Asn	Asp	Glu	Cys	Met	Glu	Ser	Val	Lys
	485				490				495						
Asn	Gly	Thr	Tyr	Asp	Tyr	Pro	Lys	Tyr	Ser	Glu	Glu	Ser	Lys	Leu	Asn
	500				505					510					
Arg	Glu	Lys	Ile	Asp	Gly	Val	Lys	Leu	Glu	Ser	Met	Gly	Val	Tyr	Asn
	515				520				525						
Ile	Leu	Ala	Ile	Tyr	Ser	Thr	Val	Ala	Ser	Ser	Leu	Val	Leu	Leu	Val
	530				535				540						
Ser	Leu	Gly	Ala	Ile	Ser	Phe	Trp	Met	Cys	Ser	Asn	Gly	Ser	Leu	Gln
	545				550				555			560			
Cys	Arg	Ile	Cys	Ile											
				565											

<210> SEQ ID NO 27  
<211> LENGTH: 1695  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: HA gene of Influenza 10-0036-2 (n+2, relative to parent)

<400> SEQUENCE: 27

atgaaagtaa	aactaatggt	tctgttatgt	acatttacag	ctacatatgc	agacacaata	60
tgtgttaggct	accatgccaa	caactcaact	gacactgttg	acacagtact	tgagaagaat	120
gtgacagtga	cacactctgt	caacctactt	gaggacagcc	acaatggaaa	actatgtcta	180
ctaaaaggaa	tagtccact	acaattgggt	aattgcagcg	ttgccccatg	gatcttagga	240
aacccagagt	gcgaattgct	gatttccaat	gaatcttgg	cctacattgt	agaaacacca	300
aatcctgaga	atggAACATG	ttaccgggg	tatTTcacag	actatgaaga	actggggag	360
caattgagtt	cagtatcttc	atTTAAGGG	ttcgaaatat	tccccaaaga	gagctcatgg	420
cccaaccaca	ccgtAAccgg	agtgtcatca	tcatgctccc	ataacgggaa	aaggcagcttc	480
tacagaaatt	tgctatggct	gacggtgaag	aacggctgt	acccaaacct	gagcaagtcc	540
tatacaaaca	aaaaggagaa	agaagtccctt	gtactatggg	gtgttcatca	cccatctaac	600
ataggggacc	aaaggccct	ctatcataca	gaaaatgctt	atgtctctgt	agtgtcttca	660
cattatacgca	gaagattcac	cccagaaata	gccaagagac	ccaagggtgag	aaatcaggaa	720
ggaagaatca	actactactg	gaccctgcta	gaaccgggg	atacaataat	atTTGAGGCA	780
aatggaaatc	taatagcacc	aaggatgcc	ttcgaactga	gtaagggttt	tggatcagga	840
atcatcacat	caaATGCACC	aatgggtgaa	tgtaatgcaa	agtgtcaaac	acctcaggaa	900

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gctataaaca gcagtcttcc tttccagaat gtacacccag taacaatagg agagtgc当地	960
aagtatgtca aaagtgc当地 attaaggatg gttacaggac taaggaacac cccatccatt	1020
caatccagag gtttggttgg agccattgcc ggtttcattg aaggagggtg gactggaatg	1080
gtagatgggtt ggtatgggtta tcaccatcg aatgagcaag gatctgggtt tgctgc当地	1140
caacaaagca cacaaaatgc cattaatggg attacaaaca aggtgaattc tgtgattgaa	1200
aaaatgaaca ctcaattcac agctgtggc当地 aaagaattca acaaaactgga aagaagaatg	1260
gaaaacttaa ataaaaaggta ttagatggg tttctagaca ttggacata taatgc当地	1320
ttgttagttc tactggaaaa tgaaaggact ttggattcc atgactccaa cgtgaagaat	1380
ctgtatgaga aagtaaaaag ccaattaaaa aataatgcc aagaaatagg aaacgggtgt	1440
tttgaattct atcataagtg taacgatgaa tgcatggaga gtgtgaaaaa ttggacttat	1500
gactatccaa aatattccga agaatcaaag ttaaacaggg agaaaattga ttggagtgaaa	1560
tttggaaatcaa tgggagttctaa taatatccctg gcgatctact caacagtccg cagttcccta	1620
gttcttttag tctccctggg ggcaatcagc ttctggatgt gttccaatgg gtctttacag	1680
tgttagaatat gc当地	1695

&lt;210&gt; SEQ ID NO 28

&lt;211&gt; LENGTH: 565

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Artificial Sequence

&lt;220&gt; FEATURE:

&lt;223&gt; OTHER INFORMATION: HA predicted AA of Influenza 10-0036-2 (n+2, relative to parent)

&lt;400&gt; SEQUENCE: 28

Met Lys Val Lys Leu Met Val Leu Leu Cys Thr Phe Thr Ala Thr Tyr			
1	5	10	15

Ala Asp Thr Ile Cys Val Gly Tyr His Ala Asn Asn Ser Thr Asp Thr		
20	25	30

Val Asp Thr Val Leu Glu Lys Asn Val Thr Val Thr His Ser Val Asn		
35	40	45

Leu Leu Glu Asp Ser His Asn Gly Lys Leu Cys Leu Leu Lys Gly Ile		
50	55	60

Ala Pro Leu Gln Leu Gly Asn Cys Ser Val Ala Gly Trp Ile Leu Gly			
65	70	75	80

Asn Pro Glu Cys Glu Leu Ile Ser Asn Glu Ser Trp Ser Tyr Ile		
85	90	95

Val Glu Thr Pro Asn Pro Glu Asn Gly Thr Cys Tyr Pro Gly Tyr Phe		
100	105	110

Thr Asp Tyr Glu Glu Leu Arg Glu Gln Leu Ser Ser Val Ser Ser Phe		
115	120	125

Lys Arg Phe Glu Ile Phe Pro Lys Glu Ser Ser Trp Pro Asn His Thr		
130	135	140

Val Thr Gly Val Ser Ser Cys Ser His Asn Gly Lys Ser Ser Phe			
145	150	155	160

Tyr Arg Asn Leu Leu Trp Leu Thr Val Lys Asn Gly Leu Tyr Pro Asn		
165	170	175

Leu Ser Lys Ser Tyr Thr Asn Lys Lys Glu Lys Glu Val Leu Val Leu		
180	185	190

Trp Gly Val His His Pro Ser Asn Ile Gly Asp Gln Arg Ala Leu Tyr		
195	200	205

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His Thr Glu Asn Ala Tyr Val Ser Val Val Ser Ser His Tyr Ser Arg  
 210 215 220  
 Arg Phe Thr Pro Glu Ile Ala Lys Arg Pro Lys Val Arg Asn Gln Glu  
 225 230 235 240  
 Gly Arg Ile Asn Tyr Tyr Trp Thr Leu Leu Glu Pro Gly Asp Thr Ile  
 245 250 255  
 Ile Phe Glu Ala Asn Gly Asn Leu Ile Ala Pro Arg Tyr Ala Phe Glu  
 260 265 270  
 Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Pro Met  
 275 280 285  
 Gly Glu Cys Asn Ala Lys Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
 290 295 300  
 Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
 305 310 315 320  
 Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
 325 330 335  
 Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
 340 345 350  
 Ile Glu Gly Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
 355 360 365  
 His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Gln Gln Ser Thr  
 370 375 380  
 Gln Asn Ala Ile Asn Gly Ile Thr Asn Lys Val Asn Ser Val Ile Glu  
 385 390 395 400  
 Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Asn Lys Leu  
 405 410 415  
 Glu Arg Arg Met Glu Asn Leu Asn Lys Val Asp Asp Gly Phe Leu  
 420 425 430  
 Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn Glu  
 435 440 445  
 Arg Thr Leu Asp Phe His Asp Ser Asn Val Lys Asn Leu Tyr Glu Lys  
 450 455 460  
 Val Lys Ser Gln Leu Lys Asn Asn Ala Lys Glu Ile Gly Asn Gly Cys  
 465 470 475 480  
 Phe Glu Phe Tyr His Lys Cys Asn Asp Glu Cys Met Glu Ser Val Lys  
 485 490 495  
 Asn Gly Thr Tyr Asp Tyr Pro Lys Tyr Ser Glu Glu Ser Lys Leu Asn  
 500 505 510  
 Arg Glu Lys Ile Asp Gly Val Lys Leu Glu Ser Met Gly Val Tyr Asn  
 515 520 525  
 Ile Leu Ala Ile Tyr Ser Thr Val Ala Ser Ser Leu Val Leu Leu Val  
 530 535 540  
 Ser Leu Gly Ala Ile Ser Phe Trp Met Cys Ser Asn Gly Ser Leu Gln  
 545 550 555 560  
 Cys Arg Ile Cys Ile  
 565

<210> SEQ ID NO 29  
 <211> LENGTH: 1695  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: HA gene of Influenza 10-0036-2 (n+1, relative  
 to parent)

<400> SEQUENCE: 29

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<210> SEQ ID NO 30
<211> LENGTH: 565
<212> TYPE: PRT
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: HA predicted AA of Influenza 10-0036-2 (n+1,
relative to parent)
```

<400> SEQUENCE: 30

Met Lys Val Lys Leu Met Val Leu Leu Cys Thr Phe Thr Ala Thr Tyr  
 1                   5                   10                   15

Ala Asp Thr Ile Cys Val Gly Tyr His Ala Asn Asn Ser Thr Asp Thr  
                  20                         25                         30

Val Asp Thr Val Leu Glu Lys Asn Val Thr Val Thr His Ser Val Asn  
35 40 45

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Leu Leu Glu Asp Ser His Asn Gly Lys Leu Cys Leu Leu Lys Gly Ile  
 50 55 60

Ala Pro Leu Gln Leu Gly Asn Cys Ser Val Ala Gly Trp Ile Leu Gly  
 65 70 75 80

Asn Pro Glu Cys Glu Leu Leu Ile Ser Lys Glu Ser Trp Ser Tyr Ile  
 85 90 95

Val Glu Thr Pro Asn Pro Glu Asn Gly Thr Cys Tyr Pro Gly Tyr Phe  
 100 105 110

Thr Asp Tyr Glu Glu Leu Arg Glu Gln Leu Ser Ser Val Ser Ser Phe  
 115 120 125

Lys Arg Phe Glu Ile Phe Pro Lys Glu Ser Ser Trp Pro Asn His Thr  
 130 135 140

Val Thr Gly Val Ser Ser Cys Ser His Asn Gly Lys Ser Ser Phe  
 145 150 155 160

Tyr Arg Asn Leu Leu Trp Leu Thr Val Lys Asn Gly Leu Tyr Pro Asn  
 165 170 175

Leu Ser Lys Ser Tyr Thr Asn Lys Lys Glu Lys Glu Val Leu Val Leu  
 180 185 190

Trp Gly Val His His Pro Ser Asn Ile Gly Asp Gln Arg Ala Leu Tyr  
 195 200 205

His Thr Glu Asn Ala Tyr Val Ser Val Val Ser Ser His Tyr Ser Arg  
 210 215 220

Arg Phe Thr Pro Glu Ile Ala Lys Arg Pro Lys Val Arg Asn Gln Glu  
 225 230 235 240

Gly Arg Ile Asn Tyr Tyr Trp Thr Leu Leu Glu Pro Gly Asp Thr Ile  
 245 250 255

Ile Phe Glu Ala Asn Gly Asn Leu Ile Ala Pro Arg Tyr Ala Phe Glu  
 260 265 270

Leu Ser Lys Gly Phe Gly Ser Gly Ile Ile Thr Ser Asn Ala Pro Met  
 275 280 285

Gly Glu Cys Asn Ala Lys Cys Gln Thr Pro Gln Gly Ala Ile Asn Ser  
 290 295 300

Ser Leu Pro Phe Gln Asn Val His Pro Val Thr Ile Gly Glu Cys Pro  
 305 310 315 320

Lys Tyr Val Lys Ser Ala Lys Leu Arg Met Val Thr Gly Leu Arg Asn  
 325 330 335

Thr Pro Ser Ile Gln Ser Arg Gly Leu Phe Gly Ala Ile Ala Gly Phe  
 340 345 350

Ile Glu Gly Gly Trp Thr Gly Met Val Asp Gly Trp Tyr Gly Tyr His  
 355 360 365

His Gln Asn Glu Gln Gly Ser Gly Tyr Ala Ala Asp Gln Gln Ser Thr  
 370 375 380

Gln Asn Ala Ile Asn Gly Ile Thr Asn Lys Val Asn Ser Val Ile Glu  
 385 390 395 400

Lys Met Asn Thr Gln Phe Thr Ala Val Gly Lys Glu Phe Asn Lys Leu  
 405 410 415

Glu Arg Arg Met Glu Asn Leu Asn Lys Lys Val Asp Asp Gly Phe Leu  
 420 425 430

Asp Ile Trp Thr Tyr Asn Ala Glu Leu Leu Val Leu Leu Glu Asn Glu  
 435 440 445

Arg Thr Leu Asp Phe His Asp Ser Asn Val Lys Asn Leu Tyr Glu Lys  
 450 455 460

Val Lys Ser Gln Leu Lys Asn Asn Ala Lys Glu Ile Gly Asn Gly Cys

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465	470	475	480												
Phe	Glu	Phe	Tyr	His	Lys	Cys	Asn	Asp	Glu	Cys	Met	Glu	Ser	Val	Lys
485															495
Asn	Gly	Thr	Tyr	Asp	Tyr	Pro	Lys	Tyr	Ser	Glu	Glu	Ser	Lys	Leu	Asn
500															510
Arg	Glu	Lys	Ile	Asp	Gly	Val	Lys	Leu	Glu	Ser	Met	Gly	Val	Tyr	Asn
515															525
Ile	Leu	Ala	Ile	Tyr	Ser	Thr	Val	Ala	Ser	Ser	Leu	Val	Leu	Leu	Val
530															540
Ser	Leu	Gly	Ala	Ile	Ser	Phe	Trp	Met	Cys	Ser	Asn	Gly	Ser	Leu	Gln
545															560
Cys	Arg	Ile	Cys	Ile											
565															

What is claimed is:

1. An attenuated swine influenza strain capable of providing a safe and effective immune response in a porcine animal against a virulent H3N2 swine influenza or diseases caused by H3N2 swine influenza;

wherein the attenuated strain encodes an HA protein having five additional glycosylation sites, relative to its corresponding virulent parental strain;

wherein the virulent parental strain encodes an HA protein having the polypeptide sequence as set forth in SEQ ID NO: 18; and

wherein the five glycosylation sites are S71N, K90N, L173T, P287T, and K294T.

2. An immunological composition comprising the strain of claim 1.

3. The attenuated strain of claim 1, containing an HA gene encoding an HA protein having the polypeptide sequence as set forth in SEQ ID NO: 22.

4. An immunological composition comprising the strain of claim 2.

5. The attenuated strain of claim 1, encoding an HA protein having only five additional glycosylation sites, relative to its corresponding virulent parental strain.

6. An immunological composition comprising the strain of claim 5.

7. The composition of claim 6, further comprising a pharmaceutically or veterinary acceptable vehicle, diluent or excipient, and which provides a protective immune response in a porcine animal against a virulent H3N2 swine influenza challenge.

8. The composition of claim 7, further comprising at least one additional antigen associated with or derived from a porcine pathogen other than H3N2 swine influenza.

9. The composition of claim 8, wherein the at least one or more additional antigen(s) is capable of eliciting in a porcine animal an immune response against *Mycoplasma hyopneumoniae* (*M. hyo*), porcine circovirus 2 (PCV2), porcine reproductive and respiratory syndrome virus (PRRSV) or other pathogen capable of infecting and causing illness or susceptibility to illness in a porcine.

10. A method of vaccinating a porcine animal in need of protection against H3N2 swine influenza, comprising administering to said porcine animal the composition of claim 6, thereby vaccinating the animal.

11. The method of claim 10, wherein the porcine animal is a sow from about 3 weeks to about 6 weeks prefarrowing.

12. The method of 11, wherein the resulting piglets have a reduced morbidity and/or mortality as compared to piglets coming from unvaccinated sows.

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